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# **EFFECTS OF FEEDING FIELD PEAS ON FRESH BEEF QUALITY**

by

Hope Voegelé

A THESIS

Presented to the Faculty of

The Graduate College at the University of Nebraska

In Partial Fulfillment of Requirements

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Major: Animal Science

Under the Supervision of Professor Chris R. Calkins

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# **EFFECTS OF FEEDING FIELD PEAS ON FRESH BEEF QUALITY**

Hope Voegelé, M.S.

University of Nebraska, 2016

Advisor: Chris Calkins

The objective of this study was to evaluate the use of field peas during two phases of production (grazing and finishing) on fresh beef quality. A total of 232 crossbred steers and heifers were randomly assigned to one of six dietary treatments in a 3x2 factorial consisting of 3 pasture and 2 finishing supplementations. The pasture phase consisted of 1) no supplement, 2) field peas at 0.5% BW, or 3) dry-rolled corn supplement at 0.5% of BW. The finishing phase consisted of 1) field peas at 20% DM or 2) no field peas.

During this two-year study, steers (year 1) and heifers (year 2), strip loin samples were aged for 14 d. Steaks were subject to retail display for a total of 7 d. Analyses included tenderness (WBSF and SSF), objective ( $L^*$ ,  $a^*$ , and  $b^*$ ) and subjective color, lipid oxidation (TBARS) and fatty acid composition. There were minimal effects due to diet. Dietary treatment had no effect on tenderness (WBSF or SSF) or content of saturated fatty acids, unsaturated fatty acids, monounsaturated fatty acids or polyunsaturated fatty acids ( $p > 0.05$ ). Although there was a significant interaction between pasture and finishing treatments for fatty acid C15:1 ( $p = 0.0331$ ), the range in values was relative low and no implications from these differences could be identified. Supplementing cattle on pasture with field peas resulted in more C18:2 ( $p = 0.0381$ ) fatty acids than when cattle were supplemented with corn, while cattle without supplement were intermediate. Meat from cattle finished with field peas had slightly greater lipid oxidation than samples

from cattle not receiving field peas during finishing (1.56 vs. 1.44 mg malonaldehyde/kg tissue, respectively;  $p = 0.0541$ ). Discoloration,  $L^*$ , and  $a^*$  displayed a triple interactions for retail display, pasture and finishing diets ( $p < 0.0001$ ,  $p = 0.0524$  and  $p = 0.024$ , respectively). Although these interactions were statistically significant, no consistent patterns could be identified. These data indicate field peas may be used for cattle with minimal negative impact on fresh meat quality.

Keyword: Beef, fatty acid composition, field peas

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## INTRODUCTION

The overall goal of this study was to determine the impacts on fresh beef quality from cattle being fed field peas (*Pisum sativum*) in a crested wheatgrass pasture production system and subsequent use in the feedlot. In recent years, field pea production has grown rapidly within the northern Great Plains. Reasons for increased production include adaptability to the climate and agronomic benefits including fixing nitrogen in soil. As the availability of field peas increase, the portion of the crop that does not meet quality standards for human consumption are being considered as an alternate nutritional feedstuff in livestock production.

Due to the nutritional value, field peas have been used as a protein supplement in ruminants (Soto-Navarro et al., 2011; Vander Pol et al., 2008, 2009) and as an energy source for monogastrics (Smith et al., 2013). Slight to no differences have been found with the inclusion of field peas on carcass quality or yield grades (Pesta et al., 2012). Researchers have also focused on tenderness differences and results indicate that dietary inclusion of field peas does not negatively impact beef palatability (Jenkins et al., 2011). To date, shelf-life studies and fatty acid profile determination of beef fed field peas has not yet been defined. Overall, there is plenty of data that has been published to determine the effects of field peas in various beef production environments but very few studies have examined the effects on fresh meats, even fewer have assessed fresh meat characteristics past tenderness determinations.

The objective of this study was to determine the impacts of field peas on meat quality. More specifically, evaluation for treatment differences on 1) Warner-Bratzler Shear Force and Slice Shear Force as indications of tenderness, 2) fatty acid profile of the

muscle, and 3) impacts on retail display and fat oxidation which impacts shelf-life and consumer acceptance of fresh beef.

## REVIEW OF LITERATURE

### I. Field Peas

Field peas (*Pisum sativum*) are rapidly becoming a significant crop in the Midwest and northern Great Plains of the United States and consequently have become an interest for both agronomists and livestock producers (Stein et al., 2004; Lardy et al., 2009). Pea cultivation and selection has led to various benefits of the crop including agronomic suitability and improved nutritional content of the seed. Annual production of field pea is used for multiple purposes including commercial, forage, rotational and cover crops (Ingels et al., 1994; Chen et al., 2006; Clark, 2007; Elzebroek and Wind, 2008). During harvest, field peas that are rejected from human consumption become a source of protein, carbohydrates, and amino acids in various livestock diets (Anderson et al., 2007).

All peas belong to the same species, *Pisum sativum*, and they are an annual cool-season legume crop that is grown in temperate climates around the world. Peas are planted in both spring and fall. “Field peas” refers to two types: winter and spring; and are also known as Austrian winter peas (black peas) and Canadian field peas, respectively (Bowman et al., 2012). The color, shape and size of the seed range in different production settings. Initially, varieties of peas were distinguished based on the purpose of the end product. According to Davies (1976), the subspecies *Arvense*, typically grown in fields, was used for animal feed and characterized by long vines, colored flowers, small pods, and dark seeds. The subspecies *Hortense*, commonly known as garden peas, were

identified with white flowers and large green or yellow seeds and were typically used for monogastric livestock or included in the human diet. Lastly, the subspecies *Axiphium*, or sugar peas, were grown for human consumption alone due to edible pods.

Peas grow best in fertile, well-drained soils with approximately 16-39 inches of annual precipitation. Pea plants grow optimally at temperatures of 12 to 17°C; however, uncovered peas can withstand temperatures as low as -10°C and if covered with snow, may tolerate temperatures as low as -30°C (Hartmann et al., 1998; Elzebroek and Wind, 2008). Winter peas, or fall-planted seeds, have a growing season of 300-320 days whereas spring peas, or spring-planted peas have a growing season of approximately 60-150 days. This includes phases from planting through flowering (Pavec, 2012).

Field peas grown as a forage crop may be grown alone or with cereals for silage and green fodder (Elzebroek and Wind, 2008). Peas can also be grazed while in the field and will regrow after being grazed multiple times (Clark, 2007). Peas and other legumes are desired as a rotational crop because they have the ability to break up disease and pest cycles, improve soil microbe diversity and activity, improve soil aggregation, and provide nitrogen (Biederbeck et al., 2005; Chen et al., 2006). Lastly, field peas are grown as green manures and cover crops especially for their nitrogen-fixing capabilities. Pea roots have nodules, formed by the bacteria *Rhizobium leguminosarum*, which converts atmospheric nitrogen ( $N_2$ ) to ammonia ( $NH_3$ ). Pea plants also produce an abundance of vines that breakdown quickly and provide additional nitrogen (Sarrantonio and Gallandt, 1994, as cited by Clark, 2007). Austrian winter peas are the most common type of pea used as a cover crop as they adapt to cold temperatures and fit in many crop rotations.

According to the Food and Agriculture Organization, over 11 million tons of dry peas were produced in 2013 globally, with the major producing countries being Canada (34%), China (14%), Russia (12%), USA (6%) and India (5%). In the United States, mixed grasses and legumes, which include field pea production, were the second largest crop commodities, only rivaled by maize production in 2013 (Pavec, 2012; USDA-NASS, 2013).

## **II. Nutrients in Field Pea**

There are no differences in total nutrient content between green and yellow peas and very subtle differences between spring- and winter-seeded peas; however, there may be small differences between some pea varieties – mainly due to differences in the size of the pea and the thickness of the hull. Mature field pea seeds consist of an outer portion, the hull, which is primarily comprised of non-starch polysaccharides. The pea kernel contains relatively high amounts of crude protein (~21-30%) and carbohydrates (~40-48%), in the form of starch, with minimal amounts of crude fat, ash and fiber (Abrahamsson et al., 1993; Castell et al., 1996). Hlovdversson (1987) found that the hull is associated with nutrient depressing factors (naturally occurring factors which interfere with nutrient availability; i.e., tannins and other enzyme inhibiting activities). The dark-flowered varieties, or the Arvense subspecies, are relatively higher in tannins and other anti-nutritional factors than their light-flowered subspecies counterparts and therefore, have lower digestibility of nutrients (Abrashamsson et al., 1993).

Field peas have a high quality amino acid profile. Peas have especially high levels of lysine and are considered a more concentrated lysine source than both cereals and

soybeans. However, peas have relatively low levels of methionine, cysteine, and sulfur amino acids (Castell et al., 1990; Hickling., 2003). In 1977, Macnicol noted the pea kernel had the ability to synthesize and inter-convert amino acids during the growing season. Albumins, globulins, and glutenins are the main protein extracts in peas. Although most non-ruminant diets must be balanced in order to meet all amino acid requirements, the amino acids in peas are highly digestible by both swine and poultry (Stein et al., 2006). The digestibility is similar or higher than in grain, and only slightly lower than in soybean meal. In ruminants, however, this protein is highly rumen degradable (Anderson et al., 2007). Amino acid content is correlated with protein content. Several factors contribute to the crude protein variability including genotypes, seed characteristics (smooth vs. wrinkled seeds), growing season, and the analytical methods (Hlodversson, 1987; Gatel and Grosjean, 1990). Peas are intermediate to cereal grains and soybeans in their crude protein content. The NRC (2001) and Hickling (2003) report an average crude protein value of 23% for peas; however, mature seeds have been reported to contain approximately 25% crude protein (Gatel and Grosjean, 1990). Spring varieties tend to have less crude protein than winter varieties (Castell et al., 1996). Additionally, wrinkled seeds are higher in average crude protein content than smooth seeds (Myer and Brendemuhl, 2001).

Like most legumes, starch is the predominant form of storage energy and is almost half of the total weight of the pea. The starch content of peas ranges from 41-54% of dry matter with approximately 50% of this being soluble. For ruminants, in high concentrate diets, the ruminal degradation rate of pea starch is similar to corn and much slower than wheat, oats, or barley. All peas, regardless of species, store starch in oval

granules. However, varietal types of peas may contain differences in the shape of the starch grains as well as the glucose polymer content: amylose and amylopectin (Grosjean, 1985; Walhain et al., 1992). Peas also contain approximately 5% oligosaccharides, made mainly of sucrose, stachyose, verbascose and raffinose. The presence of oligosaccharides represents a potential problem for swine, as pigs are deficient in galactosidase enzymes and therefore have issues in the hindgut that cause flatulence (Aman and Graham, 1987; Hickling, 2003). Also, in 1997, Igbasan et al. profiled non-starch polysaccharide content of peas, which was predominantly made of glucose, uronic acids, arabinose, xylose and galactose. The crude fiber, ash and starch content of a feedstuff influence the feed's energy and dry matter digestibility. Crude fiber levels range from 5-8%, dry matter basis, with the pea hull containing up to 75% fiber; twice as much crude fiber as corn (Coxon and Wright, 1985; Castell, 1990).

The oil, or fat, content of field peas is relatively low. The fatty acid profile of the oil in peas is similar to cereal grains, being primarily polyunsaturated. While the saturated fat content is approximately only 15%, the majority of the composition is unsaturated fatty acids (80-84%). The major unsaturated fatty acids are linoleic (45-50%), oleic (20%) and linolenic (12%) (Savage and Deo, 1989).

Field peas, like cereal grains, are low in calcium but contain a slightly higher level of phosphorus content (0.4%). Field peas also contain phytic acid (~1.2%), which binds phosphorus so that it is less available to the animal and compares favorably to soybeans (1.0-1.93%) (Hlodversson, 1987; Gatel and Grosjean, 1990). In 1998, NRC reported the amount of 0.39% phosphorus and 0.11% calcium while corn contained 0.28% and 0.03%, respectively. However, Jurgens (2002) reported the phosphorus content of pea



seed to be as high as 0.46% and Habiba (2002) reported a reduction of phosphorus availability when cooked or processed. The level of trace minerals in peas is considered to be similar to those found in cereal-based diets. There is only limited information on the vitamin content of field peas. Feeding studies did not indicate that any special or abnormal vitamin supplementation is necessary.

Similar to other legume crops, field peas contain a number of anti-nutritive factors (i.e., protease inhibitors, tannins, lectins, phytic acid, and oligosaccharides). However, in general, field peas contain very low levels of anti-nutritive factors and no special precautions are necessary before using them in animal feed. Most pulse crops (i.e., crops that include edible seeds of legumes) do contain protease inhibitors – the main ones including trypsin and chymotrypsin inhibitor. Yet again, for peas, the levels of trypsin inhibitor are low enough (usually less than 4 TIA [trypsin inhibiting activity]/mg) to be of little concern. It should be noted, however, that winter-seeded pea varieties have slightly higher levels of trypsin inhibitor (>6 TIA/mg) than spring-seeded varieties (Fan et al., 1994; Gatel, 1994). Tannins are phenolic compounds found most concentrated in the seed coat. The condensed tannins cause reduced protein and amino acid digestibility by forming indigestible linkages with protein. High tannin levels are found in brown peas, however, tannin levels are insignificant in green and yellow field peas (Griffiths, 1984). Lastly, a minor concern specifically in swine and poultry diets is the presence of oligosaccharides found in field peas. Monogastics lack alpha-galactosidase (which is primarily responsible for breaking sugars down) in the intestinal mucosa and by escaping to the large intestine forces bacterial galactosidase to break these sugars down resulting in flatulence, therefore, impairing nutrient digestion, causing cramps and general animal

discomfort. Nonetheless, field peas contain very minute levels of oligosaccharides (Hickling, 2003).

### **III. Field Peas in Ruminants**

As field peas have become a more viable feedstuff, producers and researchers alike have sought to determine the optimum inclusion level in varying stages of cattle production. Field pea protein is highly degradable in the rumen. Estimates of rumen degradability of the protein range from 78 to 94 percent, leaving modest amounts of rumen undegradable protein, also commonly referred to as by-pass protein (NRC, 1989; Aufrere et al. 1994). Lindberg (1981) found the disappearance rate of field peas within the rumen was slower during the first six hours (1.6% per hr) after consumption than for soybean meal, but increased thereafter (4.5% per hr). Lindberg concluded slower degradability protein of peas might be beneficial for rumen microbial growth and have a positive influence on rumen pH and feed efficiency. Additionally, starch is degradable in the rumen; however, starch degrades much more slowly in the rumen than wheat or barley and at nearly the same rate as corn (Walhain et al., 1992; Robinson and McQueen, 1989).

#### *Field Peas in Beef Cattle Creep Feeds.*

This feed may be used in diets where nutrient density and palatability are of importance, including creep feed diets. In 1999, Anderson reported DMI and ADG increased while G:F ratio decreased as field peas increased from 0 to 100% of the diet with optimum inclusion rates ranging from 33 to 67% of the diet. Additionally in 2004, Gelvin et al. reported calves had a greater ADG and final BW when

supplemented with field peas at 0.45% BW on a dry-matter basis. In a separate experiment, supplemented calves had greater total intake (g/kg of BW; forage + milk + creep) compared with control calves when supplemented with field peas with 8 to 16% salt in a soybean meal based diet (Gelvin et al., 2004). However, a different study found no differences in calf performance as field peas replaced up to 100% of wheat middlings (Landblomen et al., 2000). Furthermore, a study that evaluated the effect of processing field pea grains (at an inclusion rate of 40%) for creep feeds found that calves tended to have a greater daily gain than those fed ground or whole field peas (Anderson et al., 2006).

*Field Peas in Beef Cattle Receiving Diets.*

Multiple studies have been conducted to determine the effects of field peas replacing cereal grains for newly-weaned calves. In 2002, Anderson and Stoltenow conducted a study in which receiving calves were divided into the following treatment groups: 100% barley, 50% barley and 50% field peas, or 100% field peas. The study lasted 42 d and DMI and BW were evaluated after 21 d and again, at the end of the study. As field pea inclusion increased, DMI increased; however, there were no differences in daily gains and G:F slightly decreased. These results were similar to a separate experiment conducted with a field pea inclusion ranging from 0 to 56% in a barley-based diets (Anderson and Stoltenow, 2004). Additionally, Gilbery et al. (2007) examined the effects of pulse grains (field peas, lentils, or chickpea) compared to a control diet of corn and canola meal for 39 d and had similar results. Calves fed field peas, lentils or chickpeas had greater overall DMI but there were no reported differences in gain efficiency due to dietary treatment. Lentils and chickpea had the greatest ADG, however, field peas were

intermediate compared to the control treatment. All treatments were similar in CP and OM (organic matter) digestibility compared to the control diet. The effects of field pea and flaxseed at varying inclusion rates in receiving calf diets were fed in order to analyze the carryover effect on finishing performance (Landblom et al., 2007). Each year, during this 3 year study, steers were assigned to one of four pelleted receiving diets: fiber-based control diet, 12.5% flaxseed, 20% field peas or 20% field pea + 12.5% flaxseed. Unlike the previous studies, when field peas were included alone in the diet, intake did not differ between treatments but rate of gain was slower while feed efficiency was greater compared to the flaxseed treatments. Also, the blended treatment of field peas and flaxseed supplement were associated with the lowest feed cost per pound of gain. After three years, it was concluded finishing weight, ADG, and G:F did not differ between treatments. Additionally, the effect of receiving diet on carcass measurements did not effect HCW or QG. Overall, the literature has been very consistent in concluding field peas in receiving diet do not compromise the calves' health, gain or carcass quality.

#### *Field Peas in Beef Cattle Growing Diets*

Multiple studies have evaluated the use of field pea as a protein or energy source to replace portions of growing diets or supplements to cattle grazing on forage (Fendrick et al., 2005a; Pesta et al., 2012; Reed et al., 2004; Soto-Navarro et al., 2004). Two studies in particular evaluated the effects of replacing corn-based diets with field peas and had similar results. As Reed et al. (2004) replaced dry-rolled corn in a 50% concentrate diet with field peas, OM, NDF and ADF disappearance increased while DMI was not impacted, essentially providing an effective supplement while potentially decreasing the need for further protein supplementation. Similarly, Fendrick et al. (2005b) conducted a

study that replaced corn silage-based diets with field peas ranging from only 0 to 26.3% of the diet and resulted in no differences with ADG and F:G, however, DMI did increase linearly with increased field pea addition in the diet. These improvements over the corn-based diets could be contributed to the lower starch content of field peas, and furthermore, could reduce the negative association effects of starch on fiber digestion issues that are often associated with growing diets. However, Soto-Navarro et al. (2004) replaced soybean hulls, barley malt sprouts, and wheat middlings in a 45% hay diet with field peas ranging from 0 to 45% and found a decrease in DMI and OMI while DM, OM, and NDF digestibility were unaffected. Conflicting research responses to field pea inclusion in growing diets appear to be at least partially due to the variation of the basal diet, which alters other characteristics such as starch content, fiber content, CP percentages and degradability and digestibility. Additionally, binder acceptability has been evaluated while feeding DDGS and field peas combined on pasture. Pesta et al. (2012) assigned heifers to treatments that were supplemented with DDGS meal on the ground, in a bunk or a 25% field pea and 75% DDGS cube on the ground. Crude protein concentrations were equal across the treatments. The cube and bunk treatments had no differences but both performed better than the ground treatment resulting in an increase in ADG and in final BW. The researchers concluded field peas could not only be fed on the ground as a protein supplement but suggest that field peas may also be used as acceptable binder for DDGS to reduce protein supplementation waste during the grazing phase of cattle production.

#### *Field Peas in Beef Cattle Finishing Diets*

Multiple studies have utilized field peas in finishing diets in comparison to other

grains (Fendrick et al, 2005a; Lardy et al., 2009; Jenkins et al. 2011) with varying results in cattle performance. Fendrick et al. (2005a) replaced dry-rolled corn with whole field peas at 0, 20, 40 or 59% inclusion levels for 143 d. There were no significant differences observed in ADG or G:F, however, a quadratic response was observed for DMI, with intake increasing up to 40% inclusion, then decreasing at 59%, but still remained higher than cattle that were not fed field peas. Jenkins et al. (2011) replaced dry-rolled corn with only slightly different field pea inclusions of 0, 10, 20 or 30% for a 119 d finishing period but saw no differences in performance, including DMI. Lardy et al. (2009) also published the results of three experiments; two experiments replaced dry-rolled corn at 0, 10, 20, or 30% with field peas on a DM basis and the third experiment replaced corn and barley sprouts diet with field peas at 0, 18, 27, or 36% cracked field peas on a DM basis. No differences were observed on final BW, ADG, G:F, DMI or calculated  $NE_m$  or  $NE_g$ . The variation in DMI response to field peas is possibly due to variances in inclusion rate. At the lower inclusion rates, field peas fill the role of protein supplementation; while at the higher rates, field peas replace grains as a source of starch.

#### *Field Peas in Dairy Cattle Rations*

Two studies have assessed the use of field peas in dairy cattle diets and similarly to beef cattle producers, relevant research, cost and availability are the key factors that determine the feasibility of feeding field peas to dairy cows (Corbett et al., 1995; Vander Pol et al., 2008). Both studies divided Holstein cows into two groups based on stage of lactation, parity, and level of milk yield. Corbett et al. (1995) replaced a based diet of soybean meal and canola meal with approximately 25% field peas as the major source of protein and the study's duration lasted 6 months, which grain levels were adjusted

monthly based on milk yield. There was a greater milk yield for cows fed pea based concentrates in early lactation. Fat-corrected milk yield was not affected by diet regardless of lactation period. Milk fat percent was significantly higher for early and mid-lactation cows fed the pea supplement. Additionally, Vander Pol et al. (2008) replaced a control diet of soybean meal and corn grain with 15% field peas (DM basis) however, this study only lasted 70 d. DMI, milk yield, 4% fat-corrected milk yield, milk fat, milk N efficiency were not affected by diet but intake of NDF was lower and total tract apparent digestibility of starch was lower with the pea diet compared to that of the control. Other studies recorded similar results but all studies have determined field peas are an acceptable source of protein for dairy producers to use in varying lactating stages without negative consequence (Khorasani et al., 2001; Vander Pol et al., 2009).

#### *Field Peas fed in Lamb Diets*

Studies have confirmed that sheep can effectively digest field peas and that peas can support excellent performance (Lanza et al., 2003; Loe et al., 2004; Scerra et al., 2011). These studies, similarly to studies in cattle, have focused on replacing based diets with field peas in order to evaluate pea protein degradation and growth performance. Lanza et al. (2003) and Loe et al. (2004) both replaced soybean meal diets with field peas ranging from 0 to 45% with no differences among treatment groups for performance or carcass characteristics among treatment groups.

#### *Effects of Processing Field Peas.*

Rolling, ensiling, grinding, or steam flaking processing has been shown to improve metabolizable energy of various grains (Owens et al., 1997). Previously

discussed research has shown field peas to be suitable alternative to other grains, it follows that further processing of field peas would also impact their feeding value.

Birkelo et al. (2000) replaced whole corn and soybean meal with 10% dry-rolled peas. No differences in performance were observed due to either inclusion or degree of processing. Alternatingly, Anderson et al. (2006) observed a response in DMI and ADG when heifers were fed dry-rolled peas compared to whole or ground peas.

#### **IV. Field Peas in Monogastrics**

##### *Field Peas in Swine Diets*

The primary use of field peas as an alternative diet around the world is in swine diets. Pigs are able to extract a great deal of energy from peas. Additionally, field peas have a high level of lysine that is invaluable for pig growth. Lastly, peas are very palatable and since peas are a good source of energy and amino acids, they tend to displace cereal grains when used in pig feeds. Several studies have been conducted to determine the effects of including field peas in all swine production settings (Castell, 1990; Cline & Richert, 2001; Stein et al., 2004). Although conditions may vary, the results from these experiments indicate that the diet has no negative effect on the palatability of the diet or growth of the animal.

For growing pigs, field peas present a potential problem due to the trypsin inhibitor. Heat processing, via extrusion, has shown some benefit to combat the effects of this anti-nutritive component. Freire et al. (1991) conducted a study to determine the difference in performance of young pigs fed raw peas (45% spring-seeded variety) or extruded peas (30% spring-seeded). Even at these very high dietary inclusion rates, the



peas did not have enough trypsin inhibitor to cause problems for growth, as the diet had no effect on the pig's performance. Research studies that evaluated the use of raw peas in the diet have resulted in mixed results throughout literature. Stein et al. (2004) demonstrated feeding field peas (6-18%) to pigs approximately two weeks after weaning had no effect on growth performance relative to control, presumably due to a relative mature digestive system. In contrast, it has been shown that pigs fed raw peas have depressed growth performance when piglets weigh between 5-10 kg; suggesting at this weight range that a poorly developed digestive system and low levels of amylase and trypsin are more critical to piglets nutrition (Owusu-Asiedu et al. 2002). This study was further supported by Friesen et al. (2006) when raw peas depressed ADG and ADFI in all aging periods of young pigs (21-56 d) after replacing corn and soybean meal with 15-30% raw peas. The researcher suggested that other factors such as poorly developed digestive systems and low levels of amylase and trypsin (which are critical for piglets nutrition) might also influence growth performance of piglets (weigh < 20 kg).

A number of studies have shown excellent performance results when raw peas are used in growing-finishing pig diets (Bell and Keith, 1990; Stein et al., 2004, 2006). Practical inclusion levels of field peas generally range from 20-40%, depending on relative nutritional value to other available feedstuffs. Provided that diets are properly balanced, especially considering the high levels of lysine and low levels of methionine and cysteine in peas, then high levels of performance can be obtained at dietary inclusion levels of near 50%. Brand et al. (2000) replaced soybean oilcake and maize meal with field peas at 16%, 32%, 48%, and 64% resulting in increased DM intake, ADG and feed conversion efficiency up to the 48% inclusion treatment. It should be noted that all of

these traits were depressed on the highest field pea level in the diet. Stein et al. (2004) evaluated the effects of field peas in both the growing period (up to 18% field peas, as-fed basis) and the finishing period (up to 36% field pea, as fed basis). These authors concluded pig performance would not be negatively affected with the assumption that the diet is properly balanced for the digestion of lacking amino acids.

Research has also been conducted, mainly in European countries, to find the effects of including field peas in breeding swine diets – particularly gestating and lactating sow diets. Peas were supplemented during gestation (16% inclusion rate) and during lactation (24% inclusion rate) for soybean meal with no negative effects for number of pigs weaned per litter, birth weights or lactation growth rate (Gatel et al., 1988).

#### *Field Peas in Poultry Diets*

Peas provide an alternate feedstuff with a good source of protein and moderate source of energy for all classes of poultry. Similar to swine, it is of extreme importance for their diet to be balanced in order to meet nutritional requirements for amino acids. The nutrient profile of field peas seems to be of more value in laying hen diets rather than broiler diets due to the broiler's higher energy requirements. It should also be noted that in order to feed field peas to poultry, they have to be processed and reduced in size so the bird may be able to digest the intracellular starch.

Multiple studies have been conducted to determine how effective field peas are in laying hen diets. Ivusic et al. (1994) published a study in which field peas were included up to 59% of the diet and compared to soy or corn diets, the only differences observed

were a lighter yolk color and thinner egg shells with increasing levels of peas in the diet. In a separate study, Ivusic et al. substituted raw peas for wheat, after heat treatments, at inclusion rates of 0, 20, 40 and 60% had similar results (1994). The authors concluded maximum performance was attained at 40% pea inclusion level but performance then decreased at the higher level. Opposite to the study of Ivusic et al. (1994), the yolk color became darker; this could be a result of replacing wheat rather than a base cereal grain (Igbanan et al., 1997). This study also presented an earlier study looking at the same diet for broilers with finding only slight improvements in nutrient digestibility but did not find any differences in performance. Brenes et al. (1989) showed feeding field peas up to 80% in a broiler diet had no negative effects on performance as well as a slight increase in growth rate. However, it is important to note that as the rate of including field peas increased, so did the supplementation of oil, in order to balance the feeds for energy levels.

## **V. Carcass Characteristics, Meat Quality, and Sensory Attributes**

The majority of the research has observed little to no differences in carcass characteristics due to field pea inclusion in finishing diets, regardless of species. The studies that have evaluated carcass characteristics and sensory profiles over beef from cattle fed field peas have only been evaluated at only 0 to 30% of the diet with contradictory results (Anderson et al., 2007; Jenkins et al., 2014; Pesta et al., 2012). Pesta et al. (2012) reported no results for smaller inclusion rates (less than 30%) of field peas but did report an increase in HCW, dressing percentage, and 12<sup>th</sup> rib fat depth at a 30% inclusion rate. However, Jenkins et al. (2011) reported a study with no differences among treatments for HCW, LM area, fat thickness at the 12<sup>th</sup> rib, yield grade or marbling

scores. The only difference recorded in this study was a quadratic response for an increase in KPH as the inclusion of field peas also increased. Lastly, Carlin et al. (2006), cited by Anderson et al. (2007), reported no differences in carcass characteristics but reported differences in WBSF (Warner-Bratzler Shear Force), as well as sensory profiles. As field peas increased from 0 to 30%, a quadratic decrease in WBSF and a linear increase in sensory tenderness were reported. Also, an increase in juiciness and perceived tenderness was detected during the sensory panel but no overall flavor differences were reported among treatments.

A few studies have also been conducted for pork quality. Stein et al. (2006) reported no differences in pork carcass composition among treatment groups (36-66% field peas) including pH, marbling, and 10<sup>th</sup> rib backfat. The objective color scores indicated dietary field peas made the LM darker and pork chops from this treatment also had less moisture compared to other treatments. Smith et al. (2014) also reported no differences in carcass quality when field peas were added to soybean diets ranging from 7 to 30%. Skatole and indole concentrations in the backfat were quantified; there were no differences in skatole concentrations but indole concentration was linearly reduced with increasing inclusion. Both studies imply field pea inclusions in finishing diets do not negatively impact carcass quality or pork palatability.

Only one study to date was found evaluating field pea diets and their impact on poultry carcass composition and quality. Laudadio and Tufarelli (2010), evaluated field peas in the diet compared to soybean meal but reported no effect on the dressing percentage, however, total collagen and water-holding capacity increased in the pea treatment. Fatty acid analysis was also reported with an increase in polyunsaturated fatty

acid concentration in the breast and drumstick, whereas, the saturated fatty acid was similar among treatments. The authors concluded pea treatments had a positive effect on the meat quality of broilers without any negative impacts on broiler chicken performance.

Minimal literature can be found regarding meat quality in lambs and goats. However, Scerra et al. (2011) evaluated intramuscular fatty acid composition of lambs fed diets containing alternate protein sources (soybean meal, faba beans, or field peas). Meat from the field pea treatment had higher proportions of the essential fatty acids C18:2  $\omega$ -6 and C18:3  $\omega$ -3, and consequently its derivatives, C20:4  $\omega$ -6 and C20:5  $\omega$ -3 respectively, were higher in meat from animals fed the field pea treatment compared to other protein sources. Additionally, linolenic acid (C18:3  $\omega$ -3) was markedly greater in the intramuscular fat from lambs fed peas than its counterparts. The author concluded the inclusion of field peas in the diets given to finishing lambs does affect their fatty acid composition; nevertheless, these results could justify the use of field peas as a protein source without negatively impacting meat quality. To this date, fatty acid compositions have not been analyzed in other species.

## **Conclusion**

Field peas have become a practical feed ingredient in nearly for all livestock producers. Much time and energy have been invested in evaluating the replacement of high priced corn with feedstuffs that compare favorably, and compete as a competitive protein and energy source. The production of field peas, livestock production settings, optimal inclusion rates, and cost should all be considered. However, little to no research has been conducted further than the feedlot and carcass characteristics. Therefore, the

objective of this study was to determine the effects on overall fresh beef quality from cattle fed field peas during the grazing and finishing phases of production.

## **MATERIALS AND METHODS**

### ***Sampling and Experimental Design***

Crossbred steers and heifers were arranged in a 2x3 factorial design (replicated over 2 yrs) to evaluate the use of field peas during two phases of production (grazing and finishing cattle). In the factorial design, three treatments were applied during the grazing phase, and two treatments were applied during the finishing phase. During the first year, 119 crossbred steers (345 kgs) were randomly allotted to one of twelve wheatgrass pastures. Pastures were assigned to receive one of three treatments 1) no supplement, 2) field peas at 0.5% body weight (BW), or 3) dry-rolled corn supplemented at 0.5% BW. The no supplement treatment served as a negative control while the dry-rolled corn supplementation served as a positive control. During the second year, 113 crossbred heifers (249 kgs) were assigned to the same factorial design. All cattle grazed on crested wheatgrass pastures at the High Plains Agriculture Lab near Sidney, NE and were then transported to the UNL Panhandle Research Feedlot near Scottsbluff, NE. All cattle were slaughtered at Tyson Fresh Meats Inc. (Lexington, Nebraska). Cattle performance data were reported by Greenwell et al. (2016).

Carcasses were chilled for 24 h before marbling attributes (score, texture and distribution) were evaluated by a United States Department of Agriculture (USDA) beef carcass supervisor. After grading, an approximate 7.62 cm-thick slice of the anterior portion of the strip loin (*Longissimus lumborum*) was collected at the 12<sup>th</sup>/13<sup>th</sup> rib area

from each side of every carcass. All samples were vacuum packaged and transferred to the Loeffel Meat Laboratory at the University of Nebraska-Lincoln.

### ***Fabrication***

Samples were immediately fabricated upon returning to the Loeffel Meat Laboratory. All samples were deboned by hand and were cut using a slicer (SE 12D manual slicer, Bizerba, Piscataway, NJ). The samples taken from the right side of the carcass were used for tenderness evaluation and fatty acid analysis. After facing the surface on both sides ( $<0.635$  cm), a 2.54 cm steak was cut for Warner-Bratzler Shear Force (WBSF) and Slice Shear Force (SSF) testing for day 0 of retail display and the remaining portion was used for fatty acid analysis. The samples taken from the left side of the carcass were used for retail display, tenderness evaluation and oxidation. After facing the surface of both sides ( $<0.635$  cm), a 2.54 cm steak was cut for WBSF and SSF testing for day 7 of retail display and the remaining portion was used for thiobarbituric acid reactive substances (TBARS). Samples for TBARS for day 0, 4 and 7 come from one steak (1.27 cm thick) that was divided into three portions and trimmed of all subcutaneous fat. The portion for day 0 was its own steak but it also was a steak sampled for proximate analysis. The steaks that were used to evaluate fatty acid composition and oxidation were vacuum packaged using a MULTIVAC 500 (Multivac, Inc., Kansas City, MO) in Prime Source Vacuum pouches (15.24 x 25.4 cm 3 mil STD Barrier). Steaks for tenderness evaluation and retail display were packaged using an INTACT machine (Cryovac, Inc., Kansas City, MO) and placed in boxes and aged for 14 days.

### ***Retail Display***

The steaks from the left side of the carcass that were cut for retail display and tenderness, as well as TBARS, were placed into labeled foam trays (Stryo-Tech Foam Manufacturing, Denver CO, white foam tray 21.6 x 15.9 x 2.1 cm). Labels were randomly assigned to the treatments so that there would be no bias as to the treatment of each steak during retail display. These steaks were overwrapped with the same oxygen-permeable polyvinyl chloride film (PSM18, Prime Source, St. Louis, MO) and were subjected to retail display conditions. All steaks were kept in the same room with ( $2 \pm 2^{\circ}\text{C}$ ) and were exposed to continuous 1,000-1,800 lux warm white fluorescence lighting. Tenderness steaks were taken out after 7 days for SSF and WBSF evaluation. The steaks for TBARS were taken out of the retail display case at day 4 or day 7, vacuum packaged and frozen at  $-80^{\circ}\text{C}$  until analyzed.

### ***Objective and Subjective Color***

Objective color measurements were made each day for seven days at about 10:00 am each day. Measurements were obtained for CIE  $L^*$ ,  $a^*$ , and  $b^*$  values using a Minolta CR-400 colorimeter (Minolta, Osaka, Japan) set at a D65 light source and  $2^{\circ}$  observer with an 8 mm diameter measurement area. The colorimeter was calibrated daily using a white ceramic tile provided by the manufacturer, and color measures were obtained by averaging 6 readings from different areas of the steak surface. The CIE  $L^*$  measured lightness (black = 0, white = 100),  $a^*$  measured redness (red = positive values, green = negative values) and  $b^*$  measured yellowness (yellow = positive values, blue = negative values).



Percent discoloration, or subjective color, was estimated every day about the same time as objective color scoring for seven days by six graduate students during the first year and eight graduate students the second year, all of whom had previous experience with tabulating subjective color scores. Zero percent discoloration was given to a steak that had no discoloration, and as the percentages increased, so did the discoloration ratings of the steak.

### ***Tenderness Evaluation***

Steaks from the right side of the carcass were used to evaluate tenderness on day 0 of retail display while steaks from the left side of the carcass were used to measure tenderness on day 7. Steaks were removed from foam trays and an internal raw temperature and weight were recorded prior to cooking. All steaks were cooked to a target temperature of 71°C on a Belt Grill (TBG60-V3 MagiGril, MagiKitch'n Inc., Quakertown, PA). Belt grill specifications were as follows: preheat = 149°C, top heat = 163°C, bottom heat = 163°C, height of gap = 2.16 cm, and cook time was approximately 5.5 minutes. After cooking, an internal temperature and weight were recorded, and slice shear force evaluation was conducted using a Food Texture Analyzer (TMS-Pro, Food Technology Corp., Sterling, VA.) with a Slice Shear force blade. The remainder of the steak was individually bagged and stored in a cooler (maintained at  $2 \pm 2^\circ\text{C}$ ). Approximately 24 h after SSF evaluation was conducted; six (1.27 cm diameters) cores were removed with a drill press parallel to the orientation of the muscle fibers. Cores were measured using a Food Texture Analyzer (TMS-Pro, Food Technology Corp., Sterling, VA.) with a Warner-Bratzler blade.

### ***Lipid Oxidation***

Steaks in the simulated retail display condition for 0, 4, and 7 d were used to measure the oxidation status. At the end of the respective retail display period, samples were vacuum packaged and kept at -80°C. Lipid oxidation was measured by the thiobarbituric acid assay (TBA) described by Ahn et al. (1998; see appendix 3) with modifications. Fourteen ml of ddH<sub>2</sub>O and 1 mL of 10% butylated hydroxyanisole (BHA) in 90% ethanol were added to 5 g of powdered sample. After homogenizing for 15 s using a Polytron homogenizer (model CH-6010; Kinematica, Luzern, Switzerland), the homogenate was centrifuged for 3,000 x g for 5 min. One ml of homogenate was added to 2 mL of 2-thiobarbituric acid (TBA) and trichloroacetic acid (TCA) mixture (15% w/v TCA and 20 nM TBA in ddH<sub>2</sub>O) and vortexed for 5 s. The sample mixture was incubated at 70°C in a water bath for 30 min to develop color. The samples were cooled in a cold-water bath for 10 min and centrifuged at 3,000 x g for 15 min. Duplicate 200 µL of supernatant aliquots from each sample were transferred to 96-well microplates (Microtest III sterile 96-well flat-bottomed microplate; Becton Dickinson & Company, Lincoln Park, NJ) and read with a microplate spectrophotometer (Model Epoch, Biotek, Winooski, VT) at 540 nm. All 96-well microplated contained standards to calculate standard curves, and each sample was calculated as mg of malonaldehyde per kg of tissue using the standard curve from each plate.

### ***Fatty Acid Analysis***

Total lipid was extracted following the chloroform-methanol procedure of Folch et al. (1957; see appendix 4). After extraction, the lipids were converted to fatty acid methyl esters according to Morrison and Smith (1964) and Metcalfe et al. (1966).

Following dicing, each lean portion of the steak was dipped in liquid nitrogen and macerated using a Waring commercial blender (Model 51BL32, Waring Commercial, Torrington, CT). Powdered samples were stored at  $-80^{\circ}\text{C}$  until analysis. Each fatty acid was determined by gas chromatography (GC) after fat extraction from samples followed by formation of methyl esters of fatty acids. Powdered lean samples (approximately one g) were dissolved in 5 mL of 2:1 chloroform: methanol (v/v) to extract the lipid fraction after vortexing for 5 s and letting them stand for 1 h at room temperature. The homogenate was filtered three times through Watman #2 filter paper into a 13 x 150 mm screw-cap tube and the final volume was brought up to 10 mL with 2:1 chloroform: methanol mix. Afterwards, 2 mL of 0.74% KCl solution were added to the lipid extract and vortexed for 5 s to separate extracted proteins. Samples were centrifuged at  $1000 \times g$  for 5 min. Following centrifugation, the aqueous layer was aspirated and evaporated to dryness under nitrogen at  $60^{\circ}\text{C}$  (Folch et al., 1957). Then, 0.5 mL of 0.5 M NaOH in methanol was added, vortexed again for 5 s and heated for 5 min at  $100^{\circ}\text{C}$  under nitrogen for saponification. To methylate the fatty acids, 0.5 mL of boron trifluoride in 14% methanol was added and heated for 5 min at  $100^{\circ}\text{C}$  (Metcalf et al., 1966). One mL of saturated salt solution and 1 mL of hexane were added and vortexed for 5 s. Following centrifugation at  $1,000 \times g$  for 5 min, the hexane layer was removed and placed in a GC vial. All GC vials were purged with nitrogen before capping and stored at  $-80^{\circ}\text{C}$  until the sample was read on a Hewlett-Packard Gas Chromatograph (Agilent Technologies, model 5890A series, Santa Clara, CA). Total fatty acids converted to methyl esters were separated on a fused silica column (Chrompack CP-Sil 88; 0.25 mm x 100 m, Santa Clara, CA), which was placed in an oven programmed from  $140^{\circ}\text{C}$  for 10 min to  $220^{\circ}\text{C}$

at a rate of 2°C/min and held at 220°C for 20 min. Total run time was 70 min. The injector and detector were programmed to work at 270°C and 300°C, respectively. Each lipid extract was separated into fatty acids by using helium as the carrier gas at a flow rate of 1mL/min. Individual fatty acids of each sample were determined by comparison of retention times with known standards. Each standard contained fatty acids that were run through GC at the same time as a set of samples in order to provide a reference point to identify fatty acids in the sample. Each fatty acid was expressed as weight percentage value, which were relative proportions of all peaks observed by gas chromatography.

### ***Proximate Analysis***

Moisture and total fat of pulverized raw meat samples were determined. Approximately 2 g of pulverized tissue in duplicate were used to quantify moisture and ash using a LECO thermogravimetric analyzer (LECO Corporation, model 604 – 100 – 400, St. Joseph, MI). Total fat was determined as outlined by AOAC (1990) using the Soxhlet extraction procedure (See Appendix 5).

### ***Statistical Analysis***

This study was conducted with a treatment design of a 3 x 2 factorial (backgrounding diet x finishing diet) and analyzed using SAS® 9.4 package, SAS Institute, Inc., USA. Objective color and percent discoloration were analyzed for treatment main effects using the PROC GLIMMIX procedure of SAS with day as repeated measures when traits were measured over time. All other analyses were conducted with PROC GLIMMIX as well; all means were separated with the LS MEANS statement and TUKEY adjustment with an alpha level of 0.05 and tendencies were considered at an alpha level of 0.10.

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### **Effects of Feeding Field Peas on Fresh Beef Quality**

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## ABSTRACT

The objective of this study was to evaluate the use field peas during two phases of production (grazing and finishing) on fresh beef quality. Cattle ( $n = 232$ ) were randomly assigned to one of six dietary treatments in a 3x2 factorial consisting of 3 pasture and 2 finishing supplementations. The pasture phase consisted of 1) no supplement, 2) field peas at 0.5% body weight (BW), or 3) dry-rolled corn supplement at 0.5% of BW. The finishing phase consisted of 1) field peas at 20% dry-matter basis (DM) or 2) no field peas. During this two-year study, strip loin samples from steers (year 1) and heifers (year 2) were aged for 14 d. Steaks were subject to retail display (RD) for a total of 7 d. Analyses included tenderness Warner-Bratzler shear force (WBSF) and Slice Shear force (SSF), objective ( $L^*$ ,  $a^*$ , and  $b^*$ ) and subjective color, lipid oxidation (TBARS) and fatty acid composition. There were minimal effects due to diet. Discoloration,  $L^*$ , and  $a^*$  displayed a triple interaction for retail display, pasture and finishing diets ( $p < 0.0001$ ,  $p = 0.0524$ , and  $p = 0.024$ , respectively). Although these interactions were statistically significant, no consistent patterns among treatments could be identified. Dietary treatment had no effect on tenderness (WBSF or SSF). Meat from cattle finished with field peas had slightly greater lipid oxidation than samples from cattle not receiving field peas during finishing (1.56 vs. 1.44 mg malonaldehyde/kg tissue, respectively;  $p = 0.0541$ ). Content of saturated fatty acids, unsaturated fatty acids, monounsaturated fatty acids, or polyunsaturated fatty acids ( $p > 0.05$ ) was unaffected. Although there was a significant interaction between pasture and finishing treatments for C15:1 ( $p = 0.0331$ ), the range in values was relatively low and no implications from these differences could be identified. Supplementing cattle on pasture with field peas resulted in more C18:2 ( $p =$

0.0381) fatty acids than when cattle were supplemented with corn, while cattle without supplement were intermediate. These data indicate field peas may be used for cattle with minimal negative impact on fresh meat quality.

Keyword: beef, fatty acid composition, field peas

## INTRODUCTION

In recent years, field pea (*Pisum sativum*) production has grown rapidly within the northern Great Plain states. Reasons for increased production include adaptability to the climate and agronomic benefits including fixing nitrogen in soil. As the availability of field peas increase, the portion of the crop that does not meet quality standards for human consumption are being considered as an alternate nutritional feedstuff for livestock production.

Due to the nutritional value, field peas have been used as a protein supplement in ruminants (Soto-Navarro et al., 2011; Vander Pol et al., 2008, 2009) and as an energy source for monogastrics (Smith et al., 2013) with varying results. Slight to no differences have been found with the inclusion of field peas on carcass quality or yield grades (Anderson et al., 2007; Stein et al., 2014). Researchers have also focused on tenderness differences and sensory panel response and results indicate that dietary field pea inclusion does not negatively impact beef palatability (Carlin et al., 2013). To date, fatty acid profiles of beef fed field peas has not yet been determined.

The decision to utilize field pea in growing and finishing rations should be based on cost, availability, and nutrient characteristics of the ration. While previous data has indicated no negative results for production purposes from dietary inclusion, there is very

limited research that has been conducted to evaluate the effect of field peas on fresh meat quality; particularly, fresh meat quality besides yield and quality grades. Therefore, the objective of this study was to determine the impacts of field peas on meat quality, including retail display characteristics, lipid oxidation, tenderness and fatty acid profiles.

## **MATERIALS AND METHODS**

### *Cattle and Dietary Treatments*

A total of 232 crossbred cattle (replicated over 2 yrs; steers during year one, heifers during year 2) were subjected to one of three background treatments on crested wheatgrass pastures with either: 1) no supplement, 2) field peas at 0.5% BW, or 3) dry-rolled corn supplemented at 0.5% BW and one of two finishing treatments: 1) supplemented with field peas (20% on a DM basis) or 2) no peas were added to the diet. . Upon arrival in the feedlot (University of Nebraska-Lincoln Panhandle Research and Extension Center, Scottsbluff, NE), all cattle remained in their original block according to original BW block in order for randomization to be fully random. During year 1, steers were fed for approximately 119 d in the feedlot while the heifers, during year 2, were finished for 131 d.

### *Sample Collection and Fabrication*

All cattle were slaughtered at Tyson Fresh Meats Inc. (Lexington, NE). Carcasses were chilled for 24 h prior to marbling determination by a United States Department of Agriculture (USDA) beef carcass supervisor. After grading, a 7.62 cm-thick slice of the anterior portion of the strip loin (*Longissimus lumborum*) was collected at the 12<sup>th</sup>/13<sup>th</sup> rib area from each side of every carcass. All samples were vacuum packaged and transferred to the Loeffel Meat Laboratory at the University of Nebraska-Lincoln.

Samples were immediately deboned by hand and were cut using a slicer (SE 12D manual slicer, Bizerba, Piscataway, NJ). The samples taken from the right side of the carcass were used for tenderness evaluation and fatty acid analysis. After facing the surface on both sides ( $<0.635$  cm), a 2.54 cm steak was cut for Warner-Bratzler shear force and Slice Shear force testing for day 0 of retail display and the remaining portion was used for fatty acid analysis. The samples taken from the left side of the carcass were used for retail display, tenderness evaluation and oxidation. After facing the surface of both sides ( $<0.635$  cm), a 2.54 cm steak was cut for WBSF and SSF testing for day 7 of retail display and the remaining portion was used for thiobarbituric acid reactive substances (TBARS). Samples for TBARS for day 0, 4, and 7 come from one steak (1.27 cm thick) that was divided into three portions and trimmed of all subcutaneous fat. The portion for d 0 was its own steak but it also was a steak sampled for proximate analysis. The steaks that were used to evaluate fatty acid composition and oxidation were vacuum packaged using a MULTIVAC 500 (Multivac, Inc., Kansas City, MO) in Prime Source Vacuum pouches (15.24 x 25.4 cm 3 mil STD Barrier). Steaks for tenderness evaluation and retail display were packaged with an INTACT machine (Cryovac, Inc., Kansas City, MO) and placed in boxes for the aging process.

All samples were placed in boxes and aged for 14 days. After aging, steaks for visual discoloration, tenderness and lipid oxidation were placed on foam trays (21.6 x 15.9 x 2.1 cm, Styro-Tech, Denver, CO), overwrapped with oxygen permeable film (PVC-OW; PSM18, Prime Source, St. Louis, MO) and placed under RD conditions for 4 and 7 d ( $2.7^{\circ}\text{C}$  under white fluorescence lighting at 1000 to 1800 lux). Steaks used for fatty acid profile, proximate composition and 0 d RD were vacuum packaged and frozen



for further analysis (-80°C). Samples trimmed of all subcutaneous fat for proximate analysis, fatty acids, and lipid oxidation were frozen in liquid nitrogen and powdered in a metal cup blender (Model 51BL32, Waring Commercial, Torrington, CT). Powdered samples were stored at -80°C.

*Subjective Color (Visual Discoloration) and Objective Color ( $L^*$ ,  $a^*$ , and  $b^*$  values)*

Visual discoloration was assessed daily during retail display with a trained six-person panel. Panelists were provided a visual discoloration guide to use as a reference. A percentage scale was used where 0% meant no discoloration and 100% meant complete discoloration. Panelists were instructed to perform the evaluation at the same time each day to minimize variation. Samples were randomly rotated daily to minimize any possible location effects.

Objective color measurements were made each day for seven days at about 10 a.m. each day. Measurements were obtained for CIE  $L^*$ ,  $a^*$ , and  $b^*$  values using a Minolta CR-400 colorimeter (Minolta, Osaka, Japan) set at a D65 light source and 2° observer with an 8 mm diameter measurement area. The colorimeter was calibrated daily using a white ceramic tile provided by the manufacturer, and color measures were obtained by averaging 6 readings from different areas of the steak surface. The CIE  $L^*$  measured lightness (black = 0, white = 100),  $a^*$  measured redness (red = positive values, green = negative values) and  $b^*$  measured yellowness (yellow = positive values, blue = negative values).

*Tenderness Evaluation –Warner-Bratzler Shear Force (WBSF) & Slice Shear Force (SSF)*

Steaks from the right side of the carcass were used to evaluate tenderness on day 0 of retail display while steaks from the left side of the carcass were used to measure tenderness on day 7. For all steaks (never frozen), an internal raw temperature and weight were recorded. Steaks were cooked to a target temperature of 160°F on a Belt Grill (TBG60-V3 MagiGril, MagiKitch'n Inc., Quakertown, PA). Belt Grill specifications were as follows: preheat = 149°C, top heat = 163°C, bottom heat = 163°C, height of gap = 2.16 cm, and cook time was approximately 5.5 minutes. After cooking, an internal temperature and weight were recorded and slice shear force evaluation was conducted using a Food Texture Analyzer with a Slice Shear force blade. The remainder of the steak was individually bagged and stored in a cooler (maintained at 33°F). Approximately 24 hours after SSF evaluation was conducted, six cores (1/2-inch diameter) were removed parallel to the muscle fiber orientation of each steak and were measured with a Food Texture Analyzer with a Warner-Bratzler blade.

*Lipid Oxidation*

Lipid oxidation was determined with the 2-thiobarbituric acid reactive substances protocol (TBARS) as described by Ahn et al. (1998) for steaks in the stimulated retail display condition for 0, 4, and 7 d. Approximately 5 g of powdered sample were weighed into a 50 mL conical tube to which 14 mL of deionized distilled water and 1 mL of BHA (10% BHA: 90% ethanol) were added. After polytroning for 15 s, the samples were centrifuged (2,000 x g for 5 min.). One mL of the supernatant was transferred to a 15 mL conical tube and 2 mL of TBA/TCA solution (15% TCA and 20 mM TBA in deionized

distilled water) was added and vortexed before placing samples in a water bath (70°C for 30 min). After cooling, samples were centrifuged (2,000 x g for 5 min) and 200 µL of supernatant were transferred to 96-well plates. All 96-well plates had standards to calculate standard curves and ultimately mg of malonaldehyde per kg of tissue read at 540 nm.

#### *Proximate Analysis*

Proximate analysis was conducted to determine fat, moisture and ash content; with, protein content determined by the difference. Fat was quantified using the Soxhlet procedure (AOAC, 1990). Samples were measured in triplicate in Whatman #2 filter paper and fat was extracted with ether. Fat percentages were averaged per sample and used to convert fatty acid percent data to mg/100 g tissue basis. Moisture and ash were determined with a LECO thermogravimetric analyzer (LECO Corporation, Model 604-100-400, St. Joseph, MI), and samples were measured in duplicate. Moisture was determined in nitrogen atmosphere with a start temperature of 25°C and an end temperature of 130°C (~17 min ramp rate). Ash was determined in oxygen atmosphere with a start temperature of 130°C and an end temperature of 600°C (30 min ramp rate).

#### *Fatty Acid Composition*

Total lipid was extracted following the chloroform-methanol procedure of Folch et al. (1957) with modifications detailed by Morisson and Smith (1964) and Metcalfe et al. (1966). Briefly, 1 g of powdered sample was weighed into a 15 mL conical tube to which 5 mL of 2:1 chloroform:methanol was added and vortexed for 5 s. After one hour, samples at room temperature were filtered through Whatman #2 filter paper onto a 13 x 150 mm glass screw cap tube, volume was brought up to 10 mL with 2:1

chloroform:methanol and 2 mL of KCl were added and vortexed. After centrifuging samples (1,000 x g for 5 min) the top organic matter layer was aspirated off and samples were dried down completely on a heating block (60°C) under constant nitrogen purge. One half mL of 0.5 M NaOH in methanol was added, vortexed and heated (100°C) for 5 min. Then, boron trifluoride in 14% methanol (0.5 mL) was added, vortexed and heated (100°C for 5 min). Subsequently, 1 mL of saturated salt solution and 1 mL of hexane were added and samples were centrifuged (1,000 x g for 5 min). The top hexane layer was carefully pipetted into gas chromatography glass vials, nitrogen purged and lids were immediately crimped on. Chromatography was done using a Chromopack CP-Sil (0.25 mm x 100 m) column with an injector temperature of 270°C and a detector temperature of 300°C (Hewlett-Packard 6890 FID GC System; Agilent Technologies, Santa Clara, CA). The head pressure was set at 40 psi with a flow rate of 1.0 mL/min. The fatty acids were identified by their retention times in relation to known standards and the percentage of fatty acids were determined by the peak areas in the chromatograph. Values were adjusted according to percent fat and values were converted to mg/100 g tissue.

### *Statistical Analysis*

This study was conducted with a treatment design of a 3 x 2 factorial (backgrounding diet x finishing diet) and analyzed using SAS® 9.4 package, SAS Institute, Inc., USA. Objective color and percent discoloration were analyzed for treatment main effects using the PROC GLIMMIX procedure of SAS with day as repeated measures when traits were measured over time. All other analyses were conducted with PROC GLIMMIX as well; all means were separated with the LS

MEANS statement and TUKEY adjustment with an alpha level of 0.05 and tendencies were considered at an alpha level of 0.10.

## RESULTS AND DISCUSSION

### *Quality Grade Assessment*

In this study, every side from all carcasses was sampled, regardless of treatment. Due to both sides being used for various laboratory analyses, the marbling scores were collected and averaged for the overall carcass. After the marbling scores were averaged, the grade distribution was calculated for the total number of samples and were as follows: 2.2% low Prime, 11.2% high Choice, 33.5% average Choice, 42.8% low Choice and 10.3% Select. There were no quality grade differences due to dietary treatment ( $P > 0.05$ ).

### *Color Evaluation*

Discoloration had triple interactions of retail display, by pasture, by finishing diets ( $P < 0.0001$ ; Table 1). As expected, discoloration increased as retail display time increased. Studies have reported a decline in the purchasing of retail displayed beef when surface discoloration reached 20% of the surface or greater (Hood and Riordan, 1973). However, samples only reached 1.47% discoloration (overall average) by d 7 irrespective of dietary treatment during both combined years. It is concluded in the current study the magnitude of difference would require extended aging periods to visually influence the color differences perceived by consumers.  $L^*$  and  $a^*$  also displayed triple interactions of retail display, by pasture, by finishing diets ( $p = 0.0524$  and  $p = 0.024$ , Table 2 and Figure 1, respectively). Even though a significant triple interaction was found for  $L^*$ , it

should be noted the values only ranged between 43.61 and 44.99. Furthermore, all treatments acted as expected for  $a^*$  and followed a downward trend from d 0 to d 7. However, on any given day, there were no statistical differences among treatments. It is concluded the magnitude in decline of  $a^*$  varied among treatments creating a significant three- way interaction. Although there were statistically significant interactions for discoloration,  $L^*$ , and  $a^*$ , no consistent patterns due to treatments could be discerned. Evaluations of  $b^*$  displayed a main effect of retail display ( $P \leq 0.0001$ ). Similarly to  $a^*$ , all values decreased in a downward trend from d 0 to d 7.

### *Tenderness*

Tenderness (measured with WBSF and SSF) only presented differences due to retail display, showing an increase in tenderness with days of retail display ( $P < 0.0001$ ). Neither backgrounding, nor finishing treatments influenced tenderness measurements. The WBSF and SSF measurements decreased from day 0 to day 7 of retail display (3.61 vs. 2.90 and 17.92 vs. 15.26; respectively), thus indicating an increase in tenderness. A strong correlation between WBSF and SSF was observed ( $r = 0.65$ ;  $P < 0.0001$ ). These results are in disagreement with those observed by Hinkle et al. (2010) in which Warner-Bratzler shear force decreased linearly as field peas increased in the diet with the lowest shear force value occurring at the highest level of peas. Carlin et al. (2013) conducted two experiments, one with similar results to that of Hinkle et al. with an increase in tenderness while the other experiment had similar results to the current study with no differences observed. It is well known the calpain-calpastatin system has an important role in tenderization of meat (Geesink et al., 2006); therefore, the authors also found a linear decrease in calpastatin activity with increasing dietary field peas. Although there

were no observed differences in the current study, these results indicate the inclusion of field peas in the diet may alter post-mortem proteolysis and provide some explanation for the increase in tenderness observed in other studies. Because these results are not consistent among studies, it would be warranted to continue to evaluate the effects of field peas in beef cattle diets that provided further research focuses on understanding the mechanism that drives the improvement in tenderness.

#### *Lipid Oxidation*

The current study indicated meat from cattle finished with field peas had slightly greater lipid oxidation than samples from cattle not receiving field peas during finishing (1.56 vs. 1.44 mg malonaldehyde/kg tissue, respectively;  $p = 0.0541$ ). By-products are generated as a result of oxidation and are responsible for the development of off-flavors in meat products (Greene, 1969). In the current study, there was a statistically significant trend for the finishing diet, however, this is not a meaningful difference and it would require a longer period in retail display conditions in order to promote greater levels of oxidation. As expected, lipid oxidation increased over time of simulated retail display ( $d0 = 0.94$ ,  $d4 = 1.46$ , and  $d7 = 2.11$  mg malonaldehyde/kg tissue;  $P < 0.0001$ ).

#### *Proximate Analysis*

Backgrounding and finishing diets had no effect ( $P > 0.05$ ) on moisture (70.30%), protein (21.03%), fat (7.17%), or ash (1.49%) content in beef. Table 3 represents the proximate analysis for each of the dietary treatments. These results are in accordance to those reported by Domenech et al. (2014) where cattle receiving several inclusion levels of full-fat or de-oiled WDGS (wet distillers grains plus solubles) had no differences in moisture, protein, fat and ash content. Additionally, Mills et al. (1992)

conducted an experiment feeding corn silage and alfalfa hay to Holstein and crossbred steers with varying levels of protein sources of either soybean meal or fish meal and observed no differences in proximate analysis of beef.

#### *Fatty Acid Composition*

Table 3 represents the fatty acid profiles of all the dietary treatments reported in mg/100 g of tissue basis. Dietary treatment had no effect on content of saturated fatty acids, unsaturated fatty acids, monounsaturated fatty acids, or polyunsaturated fatty acids ( $P > 0.05$ ; Table 4). There was a significant interaction between pasture and finishing treatments for C15:1 but the range in values was relatively low and no implications from these differences could be identified. Supplementing cattle on pasture with field peas resulted in significantly more C18:2 fatty acids than when cattle were supplemented with corn, while cattle without supplement were intermediate. However, these differences did not carry over into total PUFA content, and did not differ among treatments. Although fatty acid composition for field peas has not been reported in beef, Scerra et al. (2011) has reported the composition of intramuscular fatty acids for lambs. Unlike the current study in which no differences were found, Scerra et al. found significant increase in linolenic acid (C18:3 n-3) and total n-3 PUFA content compared to lambs given faba bean or soybean meal, resulting in a decrease in the n-6/n-3 ratio. Furthermore, even with differences between the two studies as for which fatty acid was affected; both studies conclude no negative impacts on meat quality.

### **CONCLUSION**

Overall, there were minimal effects on color, tenderness and other shelf-life characteristics of fresh meat. In conclusion, these data indicate field peas may be used as



an alternative diet for growing and finishing cattle with minimal to no negative impact on fresh meat quality.

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**Table 1. Discoloration (%) of strip loin steaks (*L. lumbrorum*) aged for 14 d from cattle fed corn, field peas or no supplement.**

Treatment		Days on retail display							
		0	1	2	3	4	5	6	7
No Supplement on Pasture	Corn Finishing	0.00 <sup>d</sup>	0.00 <sup>d</sup>	0.00 <sup>d</sup>	0.01 <sup>d</sup>	0.01 <sup>d</sup>	0.10 <sup>d</sup>	0.41 <sup>b,c,d</sup>	0.84 <sup>a,b,c,d</sup>
No Supplement on Pasture	Field Pea Finishing	0.01 <sup>d</sup>	0.04 <sup>d</sup>	0.04 <sup>d</sup>	0.05 <sup>d</sup>	0.18 <sup>d</sup>	0.33 <sup>b,c,d</sup>	1.17 <sup>a,b,c,d</sup>	2.03 <sup>a</sup>
Field Peas on Pasture	Corn Finishing	0.00 <sup>d</sup>	0.00 <sup>d</sup>	0.00 <sup>d</sup>	0.01 <sup>d</sup>	0.11 <sup>d</sup>	0.18 <sup>d</sup>	0.86 <sup>a,b,c,d</sup>	1.62 <sup>a,b,c</sup>
Field Peas on Pasture	Field Pea Finishing	0.00 <sup>d</sup>	0.01 <sup>d</sup>	0.02 <sup>d</sup>	0.02 <sup>d</sup>	0.08 <sup>d</sup>	0.30 <sup>c,d</sup>	0.68 <sup>b,c,d</sup>	1.33 <sup>a,b,c,d</sup>
Corn on Pasture	Corn Finishing	0.03 <sup>d</sup>	0.05 <sup>d</sup>	0.07 <sup>d</sup>	0.10 <sup>d</sup>	0.16 <sup>d</sup>	0.30 <sup>c,d</sup>	0.73 <sup>a,b,c,d</sup>	1.66 <sup>a</sup>
Corn on Pasture	Field Pea Finishing	0.02 <sup>d</sup>	0.02 <sup>d</sup>	0.03 <sup>d</sup>	0.04 <sup>d</sup>	0.05 <sup>d</sup>	0.11 <sup>d</sup>	0.50 <sup>b, c, d</sup>	1.33 <sup>a,b,c,d</sup>

<sup>a-d</sup>Means with different superscripts differ ( $P \leq 0.05$ ).  $SEM = 0.3585$

**Table 2. L\* values of strip loin steaks (*L. lumbarum*) aged for 14 d from cattle fed corn, field peas or no supplement.**

Treatment		Days on retail display <sup>1</sup>							
		0	1	2	3	4	5	6	7
No Supplement on Pasture	Corn Finishing	43.69	44.99	44.46	44.47	44.48	44.51	44.76	44.69
No Supplement on Pasture	Field Pea Finishing	44.32	43.89	43.70	44.13	43.73	43.92	44.61	44.11
Field Peas on Pasture	Corn Finishing	44.49	43.86	43.72	44.14	43.86	43.83	44.04	43.71
Field Peas on Pasture	Field Pea Finishing	44.59	43.90	44.01	43.97	43.61	44.56	44.48	44.54
Corn on Pasture	Corn Finishing	44.05	43.71	43.91	44.78	43.82	44.33	44.07	44.43
Corn on Pasture	Field Pea Finishing	44.54	44.82	44.24	44.51	44.13	44.59	44.34	44.13

<sup>1</sup>There were no differences among treatments ( $P > 0.05$ ).  $SEM = 0.4909$

Treatment		Moisture	Fat	Ash	Protein <sup>2</sup>
No Supplement on Pasture	Corn Finishing	70.38	7.11	1.49	21.02
No Supplement on Pasture	Field Pea Finishing	69.82	7.73	1.43	21.02
Field Peas on Pasture	Corn Finishing	70.37	6.99	1.51	21.13
Field Peas on Pasture	Field Pea Finishing	70.60	6.92	1.54	20.94
Corn on Pasture	Corn Finishing	70.36	7.13	1.51	21.02
Corn on Pasture	Field Pea Finishing	70.26	7.13	1.49	21.12

**Table 3. Proximate analysis<sup>1</sup> of strip loin steaks (*L. lumbrorum*) aged for 14 d from cattle fed corn, field peas or no supplement.**

<sup>1</sup>There were no differences among treatments ( $P > 0.05$ ).  $SEM = 0.3610$

<sup>2</sup>Protein was calculated by subtracting the sum of moisture, fat, and ash from 100

**Table 4.** Amount<sup>1</sup> of fatty acids of beef from cattle fed corn, field peas or no supplement (*L. dorsi*)

Fatty Acid	Treatment						<i>P</i> -value			<i>SE</i> <sup>2</sup>
	No supplement on pasture		Field Peas on pasture		Corn on pasture					
	Corn Finishing	Field Peas Finishing	Corn Finishing	Field Peas Finishing	Corn Finishing	Field Peas Finishing	Pasture	Finishing	Pasture* Finishing	
C10:0	3.97	5.11	5.40	5.11	4.41	4.45	0.28	0.52	0.44	0.69
C12:0	4.20	5.56	4.70	4.87	4.53	4.35	0.69	0.30	0.31	0.61
C14:0	185.96	204.88	179.51	184.02	180.12	184.34	0.34	0.28	0.73	11.40
C14:1	44.88	49.22	40.92	45.31	42.25	44.67	0.41	0.16	0.94	3.50
C15:0	32.89	32.62	29.51	29.41	31.40	29.48	0.23	0.63	0.88	2.12
C15:1	40.40	49.64	51.02	43.43	45.72	45.02	0.76	0.90	0.03	3.48
C16:0	1,688.57	1,852.63	1,655.76	1,627.45	1,653.85	1,677.08	0.32	0.47	0.55	98.37
C16:1	240.45	271.14	241.89	254.20	251.59	260.25	0.81	0.13	0.70	15.12
C17:0	99.61	110.05	104.09	95.75	103.14	94.50	0.61	0.68	0.24	7.00
C17:1	94.98	102.52	98.79	85.78	89.13	85.12	0.21	0.55	0.29	7.10
C18:0	1,059.83	1,158.86	1,059.53	1,006.33	1,010.95	1,009.54	0.25	0.77	0.46	66.97
C18:1	2,892.93	3,209.57	2,886.72	2,801.20	2,876.43	2,973.08	0.41	0.39	0.44	169.91
C18:1v	114.01	124.05	115.85	115.97	100.62	104.09	0.16	0.54	0.86	9.98
C19:0	44.65	36.22	32.02	39.54	33.51	33.20	0.30	0.91	0.22	5.06
C18:2TT	273.28	252.67	267.17	255.87	272.13	238.71	0.93	0.20	0.87	23.96
C18:2	230.40	239.45	298.65	252.77	223.82	225.08	0.04 <sup>a</sup>	0.49	0.37	23.02
C18:3ω3	11.37	15.39	15.59	15.00	14.16	14.04	0.25	0.24	0.10	1.35
C20:0	22.94	20.97	26.18	20.64	24.52	23.83	0.47	0.06	0.37	1.95
C20:1	26.04	26.02	24.20	23.55	22.01	27.76	0.70	0.41	0.31	2.83

C20:3 $\omega$ 6	14.01	15.24	14.47	14.28	14.96	13.46	0.91	0.84	0.35	1.03
C20:4 $\omega$ 6	41.11	42.05	43.15	41.96	43.01	41.34	0.91	0.73	0.83	2.44
C22:5	12.45	14.57	11.98	12.07	12.28	11.56	0.13	0.50	0.26	0.94
Total	7,106.65	7,749.47	7,092.94	6,894.24	7,000.84	7,059.18	0.41	0.57	0.49	387.66
Other	22.77	49.72	46.15	27.79	52.25	50.93	0.32	0.80	0.14	12.48
SFA <sup>2</sup>	3,123.64	3,372.91	3,075.65	2,994.65	3,024.53	3,040.94	0.33	0.65	0.60	179.91
UFA <sup>2</sup>	3,983.02	4,376.57	4,017.30	3,899.59	3,976.32	4,018.24	0.49	0.51	0.42	214.44
SFA:UFA <sup>2</sup>	0.78	0.78	0.77	0.77	0.77	0.76	0.54	0.69	0.95	0.02
MUFA <sup>2</sup>	3,433.64	3,817.86	3,429.83	3,358.83	3,419.25	3,505.81	0.44	0.38	0.46	199.74
PUFA <sup>2</sup>	549.38	558.70	587.47	540.77	557.07	512.43	0.60	0.26	0.57	32.44
$\omega$ 6	52.56	52.93	54.63	54.21	55.24	51.30	0.83	0.57	0.72	3.07
$\omega$ 3	11.37	15.39	15.59	15.00	14.16	14.04	0.25	0.24	0.10	1.35
$\omega$ 6: $\omega$ 3	4.59	3.90	3.96	3.81	4.66	4.00	0.34	0.06	0.64	0.38

<sup>1</sup>Amount (mg/100 g tissue) of fatty acid in powdered loin sample determined by gas chromatography. Fat percentage was calculated by proximate analysis.

<sup>2</sup>SEM = Standard Error of the Mean, SFA = Saturated fatty acids, UFA= Unsaturated fatty acids, SFA:UFA = Saturated fatty acids: Unsaturated fatty acids, MUFA= Monounsaturated fatty acids, and PUFA=Polyunsaturated fatty acids

<sup>a</sup>For C18:2, means separation were as follows: no supplement = 234.93<sup>ab</sup>, field pea = 275.71<sup>a</sup>, and corn = 224.45<sup>b</sup>.

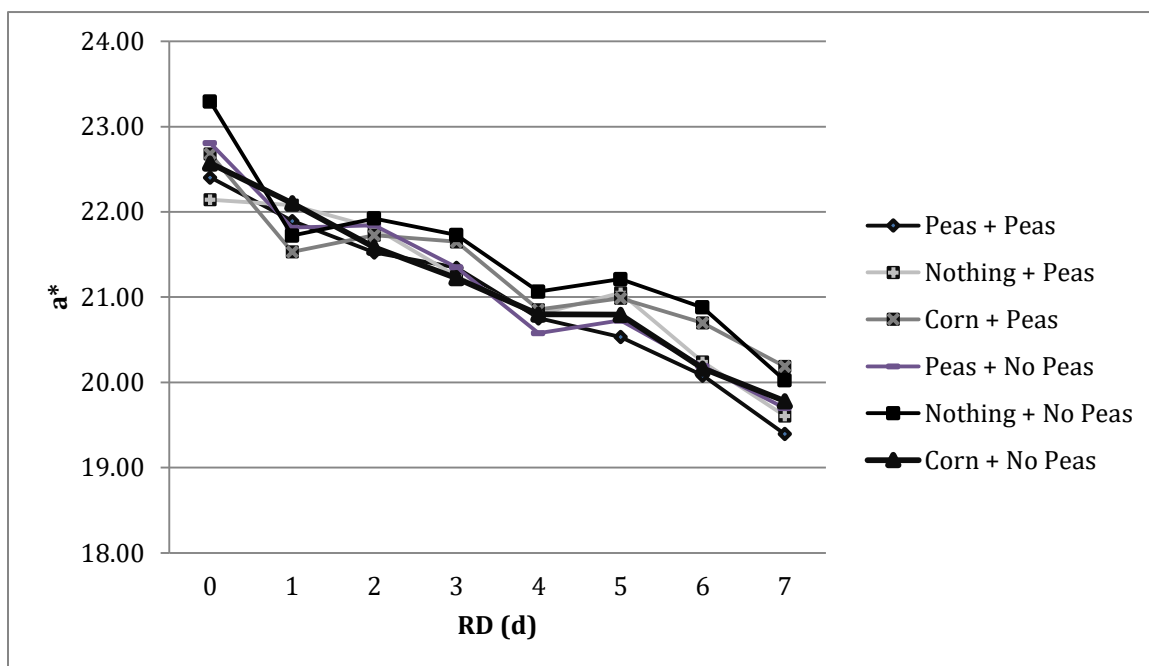


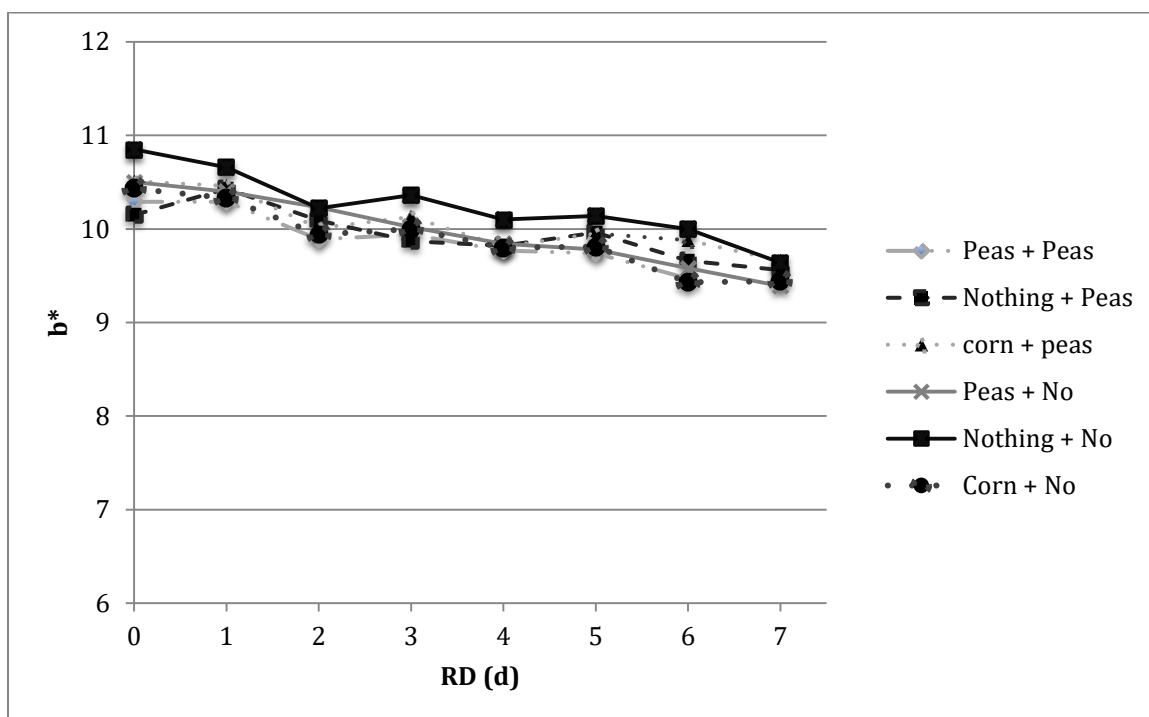
**Figure 1.**  $a^*$  values of strip loin steaks (*L. lumbarum*) aged for 14 d from cattle fed corn, field peas, or no supplement.

Retail display by pasture treatment by finishing treatment interaction ( $p = 0.0240$ ;  $SEM = 0.3747$ )

**Figure 2.**  $b^*$  values of strip loin steaks (*L. lumbarum*) aged for 14 d from cattle fed corn, field peas, or no supplement.

$b^*$  values displayed a retail display effect ( $P < 0.0001$ ;  $SEM = 0.0690$ )





## **RECOMMENDATIONS FOR FUTURE RESEARCH**

Although the current study had few significant differences, I believe there are many possibilities to build upon my work and others in literature. Some suggestions for future research can include:

1. The current study evaluated field peas vs. other dietary treatments so rather than a black or white study, future research may analyze the same fresh meat qualities in varying percentages in the diet. True, this has been well established in research in multiple production settings but fresh meat evaluation has not.
2. This study established the fatty acid profile of lean samples but it may be interesting to establish the fatty acid profile of the subcutaneous fat. Although very minimal discoloration was seen for the lean portion of the steak, it was noted the fat portion of the steak not only became darker, but also looked slightly green during the subjective color analysis. Depending on the fatty acids present, it may explain the lack of differences in color and lipid oxidation.
3. I would have also liked to extend the retail display portion of this study. Subjective discoloration peaked at 1.47% (overall average) across all treatments but I am unsure if this trend would continue if the retail display portion was extended to 10 days or more.
4. There are still very large differences in tenderness that have been reported for beef fed field peas. I believe more differences would be found if the inclusion level was varied.
5. To date and to the authors knowledge, only one study has evaluated the consumer acceptance of beef fed field peas and in that study, there were positive

correlations between the increase inclusion of field peas and juiciness and tenderness identified by a consumer panel. If this trend was to continue and consumers could identify differences for treatments, it may be of value to validate a new study.

## Appendix I. DIETARY TREATMENT (Year 1 & 2)

### Grazing Phase:

1. Control Treatment: No supplement, solely grazed pasture
2. Field Pea Treatment: Supplemented at 0.5% BW, dry matter basis
  - 100% Field Peas
3. Corn Treatment: Supplemented at 0.5% BW, dry matter basis
  - 70.8% Dry-rolled corn, 24% Solubles, 5.2% Urea
4. Pastures nutrient composition is below:

Table 1. Nutrient Analysis of Pastures 2015 – Year 2

Nutrient Analysis	June 2015	August 2015	SEM	P-value
Organic Matter	89.0%	88.9%	0.367	0.866
IVDMD	49.0%	40.3%	1.018	0.004
IVOMD	48.2%	38.8%	--	--
NDF	69.5%	68.8%	0.854	0.623
ADF	47.6%	48.0%	0.305	0.399
CP	8.84%	6.39%	--	--

### Finishing Phase:

Cattle were fed *ad libitum* during the entire feeding period. Two treatments and composition are listed below. Percentages are on a dry matter basis. Tylan (90 mg/hd) and Rumensin (360 mg/hd/day) were also provided in the diet.

Feedstuff	"No Peas"	"Peas"
DRC	60%	40%
WDGS	20%	20%
Field Peas	0%	20%
Corn Silage	14%	14%
Supplement	6%	6%

## Appendix II. NUMBER OF CATTLE PER TREATMENT

Treatment Identification		
Assigned Code	Pasture Treatment	Feedlot Treatment
TRT A	Pea supplement (0.5% BW)	Peas added (20% DM basis)
TRT B	No supplement	Peas added (20% DM basis)
TRT C	Corn supplement (0.5% BW)	Peas added (20% DM basis)
TRT D	Pea supplement (0.5% BW)	No peas added
TRT E	No supplement	No peas added
TRT F	Corn supplement (0.5% BW)	No peas added

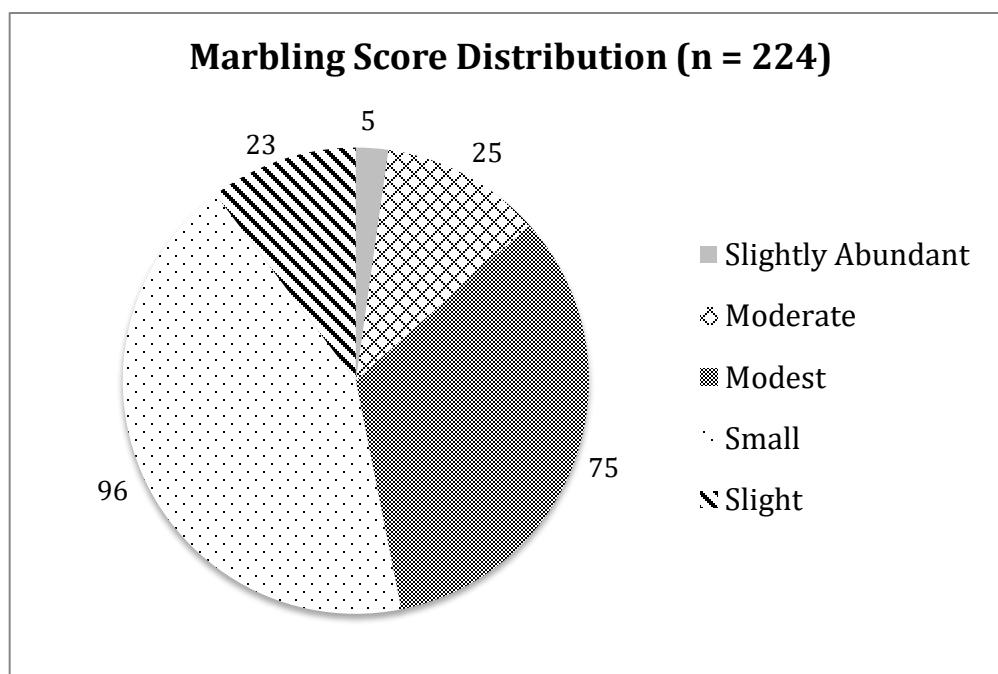
Crossbred steers and heifers were arranged in a 2x3 factorial design (replicated over 2 yrs) to evaluate the use of field peas during two phases of production (grazing and finishing cattle). In the factorial design, three treatments were applied during the grazing phase, and two treatments were applied during the finishing phase. Cattle were randomly allotted to one of twelve crested wheatgrass pastures. Pastures were assigned to receive one of three treatments 1) no supplement, 2) field peas at 0.5% body weight (BW), or 3) dry-rolled corn supplemented at 0.5% BW. The no supplement treatment served as a negative control while the dry-rolled corn supplementation served as a positive control.

Treatment	Year 1	Year 2
A	20	20
B	20	20
C	16	17
D	18	18
E	18	18
F	19	20
Total	111	113

During the first year, 119 crossbred steers (345kg) were fed during the study; however, analyses on steaks were only conducted on 111 samples. Reasons behind this difference included U.S. Retained carcasses, calloused muscle, etc. During the second year, 113 crossbred heifers (249 kg) were assigned to the same factorial design and all of these steaks were collected for analysis. Laboratory analyses were conducted on 224 steaks.

### Appendix III. QUALITY GRADE ASSESSMENT

In this study, every side from all carcasses was collected regardless of treatment. Due to both sides being used for various laboratory analyses, the marbling scores were collected and averaged for the overall carcass.



After the marbling scores were averaged, the marbling grade distribution was calculated for the total number of head in the study. The grade distribution ranged from low prime to low select. There were no quality grade differences due to dietary treatment ( $P > 0.05$ ).

Quality Grade	Cattle Distribution (%)
Low Prime	2.2
High Choice	11.2
Average Choice	33.5
Low Choice	42.8
Select	10.3



## Appendix IV. COLOR AND DISCOLORATION

### OBJECTIVE COLOR - Minolta Calibration Procedures

#### 1. Before Calibration:

Calibrate Minolta in the same temperature conditions as the measurements being taken.

- Place the Minolta in the environment where samples will be measured about 5 or 10 minutes before calibrating so it can become equilibrated with the temperature.

Calibrate with the same materials as you will be taking measurements.

- If the measurements will not be taken directly on the meat surface, you must calibrate the Minolta with the same material it will be measuring through. For example, if you want to take readings from samples that are wrapped in overwrap, you must put some overwrap around the measuring head “eye” while calibrating using the white tile.

2. Turn the power to the measuring head *ON*.

3. Turn the power to the data processor *ON* while holding down the [DELETE/UNDO] key at the same time.

- Release the [DELETE/UNDO] key when you hear a “beep”.

(This deletes any previous data that might still be stored in the data processor)

4. When the screen turns on, the question “Initial set ok?” appears, press the [Measure Enter] key.

5. Once you get to the measurement screen, press the [Index Set] key.

- Use the *arrows and the [Measure Enter] key* to adjust all the following settings:
  - Printer → On
  - Color space → Off
  - Protect → On
  - Auto Average → However many readings wanted per sample (1-30)
  - Illuminant → D65
  - Back light → Off
  - Buzzer → On
  - Disp. Limit

- Press the [Esc] key to return to the measurement screen.

6. Press the [Calibrate] key while in the measurement screen.

7. Enter in the numbers listed on the calibrating white tile for the D65 setting using the following:

- [< >] keys and the numeric pad
- (The [< >] keys move the cursor)
- D65 settings: Y: 93.13  
                                   x: 0.3164  
                                   y: 0.3330

8. Set up the measuring head so that it is resting on the LCD screen and the “eye” is facing up.

Place the white calibration tile on the measuring head, near the middle of the tile.

9. Press either the measurement button on the measuring head OR the [Measure Enter] key on the data processor after making sure the ready lamp is ON.

- Make sure the white tile is completely on the measuring head “eye”.
- The Calibration is complete after the lamp flashes 3 times and the screen returns to the measurement screen.
- Do not move the measuring head during calibration.

10. Press the [Color Space] key until the L\*, a\*, b\* screen shows up.

11. Calibration is finished and the Minolta is ready

- To save battery life, turn both the measurer and data collector off after calibration is finished until you need it for measuring. The calibration and settings will not be erased.
- When turning back on for measurements, ONLY turn on the power buttons. DO NOT hold down the [DELETE/UNDO] key at the same time. This will delete the calibration and settings and all of the steps will have to be repeated.

### Cleaning

- Wipe machine down with a soft, clean dry cloth. Never use solvents such as thinner or benzene.
- If the white calibration tile becomes dirty, wipe it gently with a soft, clean dry cloth. If dirt is difficult to remove, wipe it with lens cleaner and cloth, then dry.

### Helpful Hints

1. Make a separate data sheet

- The Minolta prints out data with sample numbers 1 to 2000. In order to correlate it back to a sample, you must make a separate data sheet that has a place to record meat sample i.d. and its corresponding Minolta number.

2. Batteries

- The measuring head requires 4 AAA batteries and the data processor requires 4 AA batteries

3. The auto protect setting

- The Minolta can only record and store up to 2000 readings, once you go past 2000 readings it will start deleting older readings.
- When the auto protect is on it will automatically prevent the 2001<sup>st</sup> reading from being taken so you cannot accidentally overwrite other data.

4. Auto Average Function

- During calibration, if you set the Auto Average function to a reading number above 1, for example 5, you only have to hit the measure button once and it will automatically take all 5 readings then print out the average.
- It only allows a second or two between readings so make sure you are paying attention and move the measuring head to where you want it before it automatically takes the next reading.

### 5. Recalibrate regularly

- If using the Minolta all day, or for long periods of time, make sure to recalibrate it regularly.

### 6. DELETE/UNDO KEY

- If you accidentally take a reading, hitting the [DELETE/UNDO] key will delete the last reading.
- If you accidentally delete a reading by hitting the [DELETE/UNDO] key, hitting the [DELETE/UNDO] key again will restore the previous reading.

### 7. Printer Paper

- The paper that the data is printed on is sensitive to heat and light. The printed data should be kept in a dark cool place, like a desk drawer. In order to prevent losing any data, you must make a photocopy of the printout in order to preserve it for long-term storage.

### 8. More than One Color Space on Print Out

If you want to print more than 1 color space (Example: L\*a\*b\* AND XYZ) on the print out slip:

1. Press the [Index Set] key. Use the arrows and the [Measure Enter] key to adjust all the following settings:
  - Color space → On
  - Disp. Limit → press the [Measure Enter] key to select this option
  - Once inside the Disp. Limit option, go through the list and change all the color spaces that you DO NOT want to OFF.
2. Press the [Esc] key until you return to the measurement screen.

### 9. Change Measurements to a Different Color Space

If you get done measuring and realize that you meant to measure in a different color space (For example: measured everything using Yxy and meant to use L\*a\*b\*), you can correct it using these steps:

1. While in the measurement screen, press the [Color Space] key until your desired color space (in this example: L\*a\*b\*) appears.
2. Press the [Data List] key while in the measurement screen.
3. Select the desired page using the up and down arrows.
  - If you only have one page it will show up as P00, select this one.
4. Once you have the desired page selected, press the [Measure Enter] key.
5. Press the [Print/Feed] key
6. Select "All Meas. Data" using the up and down arrows.
7. Press the [Measure Enter] key
  - This will reprint all the stored data in your newly selected color space (L\*a\*b\* in this example).
8. Press the [Esc] key to return to the measurement screen.

SUBJECTIVE COLOR –Percent Discoloration Chart**0%****5%****10%****20%****30%****40%****50%****60%****70%****80%****90%****100%**

## Appendix V. THIOBARBITURIC ACID REACTIVE SUBSTANCES ASSAY

(Buege and Aust, 1978), Modified by Ahn et al., (1998)

### Standards:

#### **TEP solution (1, 1, 3, 3-Tetraethoxypropane)** (Make new weekly)

Stock Solution: Dilute 99 $\mu$ l TEP (97%) bring volume to 100 mL ddH<sub>2</sub>O

Working Solution: Dilute stock solution to 1:3 (TEP Solution:ddH<sub>2</sub>O) ( $1 \times 10^{-3}$ M)

#### **TBA/TCA (2-Thiobarbituric Acid/Trichloroacetic Acid) Stock Solution: 1L**

15% TCA (w/v) and 20 mM TBA (MW 144.5) reagent in ddH<sub>2</sub>O

Dissolve 2.88 g TBA in warm ddH<sub>2</sub>O first, then add TCA (150g) and ddH<sub>2</sub>O to 1L

#### **BHA (Butylated HydroxyAnisole) Stock Solution:**

Make 10% stock solution by dissolving in 90% ethanol

10g BHA dissolved in 90mL ethanol (90%) + 5mL ddH<sub>2</sub>O

### **Standards: In Duplicate**

		<u><b>Moles of TEP</b></u>
Blank:	1 ml ddH <sub>2</sub> O	
Standard 5:	100 $\mu$ l working TEP + 1.90 mL ddH <sub>2</sub> O	( $5 \times 10^{-5}$ M)
Standard 4:	1 mL Std. 5 + 1 mL ddH <sub>2</sub> O	( $2.5 \times 10^{-5}$ M)
Standard 3:	1 mL Std. 4 + 1 mL ddH <sub>2</sub> O	( $1.25 \times 10^{-5}$ M)
Standard 2:	1 mL Std. 3 + 1 mL ddH <sub>2</sub> O	( $0.625 \times 10^{-5}$ M)
Standard 1:	1 mL Std. 2 + 1 mL ddH <sub>2</sub> O	( $0.3125 \times 10^{-5}$ M)

Remove 1 mL of Standard 1 and discard it, leaving 1 mL behind.

TBA Procedure:

1. Mix all reagents and standards before beginning.
2. Transfer 5g powdered sample into a 50mL conical tube; add 14 mL of ddH<sub>2</sub>O and 1.0 mL of BHA.
3. Homogenize for 15 sec with a polytron.
4. Centrifuge for 2000xg for 5 min.
5. Transfer 1mL of homogenate or standard to 15mL conical tube.
6. Add 2 mL of TBA/TCA solution, vortex.
7. Incubate in a 70°C water bath for 30 min. to develop color.
8. Cool samples in a cold water bath for 10 min.
9. Centrifuge tubes at 2000xg for 15 min.
10. Transfer duplicate aliquots of 200 µl from each tube into wells on a 96-well plate.
11. Read absorbance at 540 nm.

Calculations: mg of malonaldehyde/kg of tissue

$$K (\text{extraction}) = (S/A) \times MW \times (10^6/E) \times 100$$

Where:

S = Standard concentration (1 x 10<sup>-8</sup> moles 1, 1, 3,3-tetraethoxypropane)/5ml

A= Absorbance of standard

MW = MW of malonaldehyde

E = sample equivalent

P = percent recovery

Final Calculation: .012 x concentration x 72.063 x 10<sup>-6</sup> = mg of Malonaldehyde/kg tissue

Reagents (Sigma): TBA – T5500; TCA – T9159; TEP – T9889; BHA – B1253

## Appendix VI. FATTY ACID DETERMINATION

Weigh out 1 g of pulverized muscle tissue. If extracting subcutaneous fat, weigh out 0.1 g of pulverized subcutaneous fat into centrifuge tube.

1. Add 5 mL of 2:1 chloroform:methanol (v/v) for muscle tissue or 3 mL for subcutaneous fat.
2. Vortex for 5 s and let stand for 1 h at room temperature.
3. Filter homogenate through Watman #2 filter paper into 13 x 150 mm screw cap tube bringing the final volume with chloroform:methanol to 10mL for muscle lipid and 5mL for subcutaneous fat extract. If stopping at this point, purge test tube with nitrogen, cap tube, and store at -80°C.
4. Add 2 mL of a 0.74% KCl solution for muscle lipid extract or 1 mL for subcutaneous fat tissue extract and vortex for 5 s. If stopping at this point, purge test tube with nitrogen, cap tube, and store at -80°C.
5. Centrifuge samples at 1000 x g for 5 min. Following centrifugation, aspirate off the aqueous phase (top layer). If stopping at this point, purge test tube with nitrogen, cap tube, and store at -80°C.
6. Evaporate to dryness under nitrogen at 60°C.
7. Add 0.5 mL of a 0.5 M NaOH in methanol. Vortex for 5 sec. Heat for 5 min at 100°C.
8. Add 0.5 mL of boron trifluoride in 14% methanol. Vortex for sec. Heat for min at 100°C.
9. Add 1 mL of saturated salt solution and 1 mL of hexane. Vortex for 5 sec.
10. Centrifuge samples at 1000 x g for 5 min. Following centrifugation, remove hexane layer (top layer) making sure not to disrupt the aqueous phase (lower layer) and place in GC vial. Purge GC vial with nitrogen, cap and crimp cap, and store at -80°C until sample is ready to be read on the GC.

### GC Settings

Column – Chropack CP-Sil 88 (0.25 mm x 100 m)

Injector Temp. - 270°C

Detector Temp. - 300°C

Head Pressure – 40 psi

Flow Rate – 1.0mL/min

Temperature Program – Start at 140°C and hold for 10 min. Following 10 min, raise temperature 2°C/min until temperature reaches 220°C. At 220°C, hold for 20 min.

## Appendix VII. FAT EXTRACTION (SOXHLET METHOD)

**WARNING: ETHER IS EXTREMELY FLAMMABLE AND PRODUCES EXPLOSIVE PEROXIDES. NEVER BRING A RADIO OR ANY OTHER POTENTIALLY SPARK-PRODUCING ITEM INTO THE FAT EXTRACTION ROOM.**

1. Check ground glass connections. They should be wiped clean with a dry paper towel and given a thin coating of stopcock grease.
2. Each boiling flask must contain boiling stones. This helps prevent violent boiling of the solvent which could be dangerous.
3. Load samples into soxhlet tubes, arranging them so that no samples are above the level of the top bend in the narrower tubing on the outside of the soxhlet. (The soxhlet will only fill the solvent up to this point before cycling back down into the boiling flask.) In general, the large soxhlets will hold about 20 two-gram samples and the small soxhlets from 4-6.
4. Fill the large (500mL) boiling flasks with @ 400 ml of solvent and the small (125mL) flasks with 100 mL of solvent. **DO THIS UNDER THE FUME HOOD!**
5. Fit the soxhlet onto the boiling flask. Very carefully, bring the assembly into the extraction room and fit it onto the condenser. Make sure all ground glass connections are snug and each boiling flask is resting on the heating element. The ceramic fiber sheet should be covering the bare metal surfaces of the burners completely.
6. Turn on the water supply to the condensers (usually a quarter turn). Check later to make sure condensers are cool enough – if not, increase water flow.
7. Turn heating element control dials to 2 way between three and four. Each burner has its own dial. **NEVER TURN THE BURNER BEYOND FIVE.** Ether has a very low boiling point and violent boiling is dangerous. Double check fittings, boiling stones, etc.
8. Fat extraction will take from 24-72 hours depending on the sample (beef – 48 hours, bacon – 72 hours). Check extractions twice daily to see that everything is alright while they are running.
9. When done, turn off the burners and let solvent cool completely before removing samples.
10. After it has cooled down, slowly uncouple the flask and soxhlet tube from the condenser. Cover the top of the soxhlet with one palm so as to reduce ether vapors while transporting it to the fume hood. Air-dry samples in the fume hood for two hours to get rid of the reaming ether in the samples. Pour ether back slowly into an approved container for reuse or discarding. **DO NOT LEAVE ETHER OUT OF THE HOOD OR THE FLAMMABLE CABINET.**
11. Place samples in drying oven (105°C) for about four hours or overnight before weighing back.

Calculation: (((Original weight including filter paper and paper clip – Fat extracted sample weight)/Sample Wt)\*100) - % Moisture = % Fat



**APPENDIX VIII. a\* values of strip loin steaks (*L. lumbrorum*) aged 14 d from cattle fed corn, field peas or no supplement.**

Treatment		Days on retail display							
		0	1	2	3	4	5	6	7
No Supplement on Pasture	Corn	23.29 <sup>a</sup>	21.72 <sup>b,c,d,e,f</sup>	21.92 <sup>a,b,c,d,e,f</sup>	21.73 <sup>b,c,d,e,f</sup>	21.07 <sup>d,e,f,g,h,i,j,k</sup>	21.21 <sup>c,d,e,f,g,h,i,j,k</sup>	20.88 <sup>e,f,g,h,i,j,k,l,m</sup>	20.02 <sup>j,k,l,m,n</sup>
No Supplement on Pasture	Field Pea	22.14 <sup>a,b,c,d,e</sup>	22.08 <sup>a,b,c,d,e</sup>	21.81 <sup>b,c,d,e,f</sup>	21.24 <sup>c,d,e,f,g,h,i,j</sup>	20.80 <sup>e,f,g,h,i,j,k,l,m</sup>	21.05 <sup>d,e,f,g,h,i,j,k,l</sup>	20.24 <sup>g,h,i,j,k,l,m,n</sup>	19.61 <sup>m,n</sup>
Field Peas on Pasture	Corn	22.81 <sup>a,b</sup>	21.82 <sup>b,c,d,e,f</sup>	21.84 <sup>a,b,c,d,e,f</sup>	21.35 <sup>c,d,e,f,g,h,i,j</sup>	20.58 <sup>f,g,h,i,j,k,l,m,n</sup>	20.73 <sup>e,f,g,h,i,j,k,l,m,n</sup>	20.19 <sup>g,h,i,j,k,l,m,n</sup>	19.70 <sup>l,m,n</sup>
Field Peas on Pasture	Field Pea	22.40 <sup>a,b,c,d</sup>	21.89 <sup>a,b,c,d,e,f</sup>	21.52 <sup>b,c,d,e,f,g,h,i</sup>	21.35 <sup>c,d,e,f,g,h,i,j</sup>	20.75 <sup>e,f,g,h,i,j,k,l,m</sup>	20.53 <sup>f,g,h,i,j,k,l,m,n</sup>	20.08 <sup>j,k,l,m,n</sup>	19.40 <sup>n</sup>
Corn on Pasture	Corn	22.57 <sup>a,b,c</sup>	22.10 <sup>a,b,c,d,e</sup>	21.58 <sup>b,c,d,e,f,g,h</sup>	21.22 <sup>c,d,e,f,g,h,i,j</sup>	20.80 <sup>e,f,g,h,i,j,k,l,m</sup>	20.79 <sup>e,f,g,h,i,j,k,l,m</sup>	20.16 <sup>i,j,k,l,m,n</sup>	19.78 <sup>k,l,m,n</sup>
Corn on Pasture	Field Pea	22.68 <sup>a,b,c</sup>	21.53 <sup>b,c,d,e,f,g,h,i</sup>	21.73 <sup>b,c,d,e,f,g</sup>	21.65 <sup>b,c,d,e,f,g</sup>	20.85 <sup>e,f,g,h,i,j,k,l,m</sup>	20.99 <sup>d,e,f,g,h,i,j,k,l,m</sup>	20.70 <sup>e,f,g,h,i,j,k,l,m,n</sup>	20.19 <sup>h,i,j,k,l,m,n</sup>

<sup>a-m</sup>Means with different superscripts differ ( $P \leq 0.05$ ).  $SEM = 0.3747$

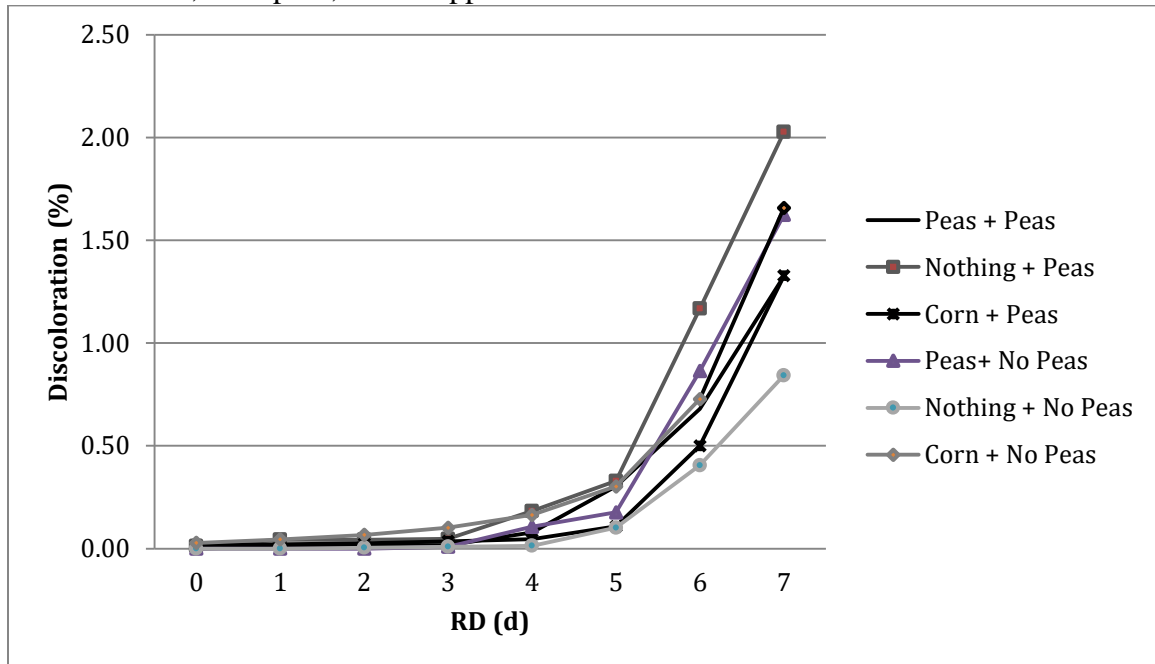
**APPENDIX IX. b\* values of strip loin steaks (*L. lumorum*) aged for 14 d from cattle fed corn, field peas or no supplement.**

Treatment		Days on retail display <sup>1</sup>							
		0	1	2	3	4	5	6	7
No Supplement on Pasture	Corn Finishing	10.85	10.66	10.22	10.36	10.10	10.14	10.00	9.64
No Supplement on Pasture	Field Pea Finishing	10.15	10.43	10.09	9.87	9.82	9.96	9.66	9.56
Field Peas on Pasture	Corn Finishing	10.50	10.40	10.23	10.02	9.84	9.78	9.58	9.39
Field Peas on Pasture	Field Pea Finishing	10.29	10.29	9.89	9.94	9.77	9.74	9.47	9.21
Corn on Pasture	Corn Finishing	10.43	10.33	9.94	10.00	9.80	9.81	9.43	9.44
Corn on Pasture	Field Pea Finishing	10.51	10.46	10.01	10.13	9.81	9.95	9.88	9.66
Overall Means <sup>1</sup>		10.46 <sup>a</sup>	10.43 <sup>a</sup>	10.06 <sup>b</sup>	10.05 <sup>b</sup>	9.86 <sup>cd</sup>	9.90 <sup>cb</sup>	9.67 <sup>d</sup>	9.48 <sup>e</sup>

<sup>1</sup> b\* values displayed a significant retail display effect ( $P < 0.0001$ ).  $SEM = 0.05891$ .

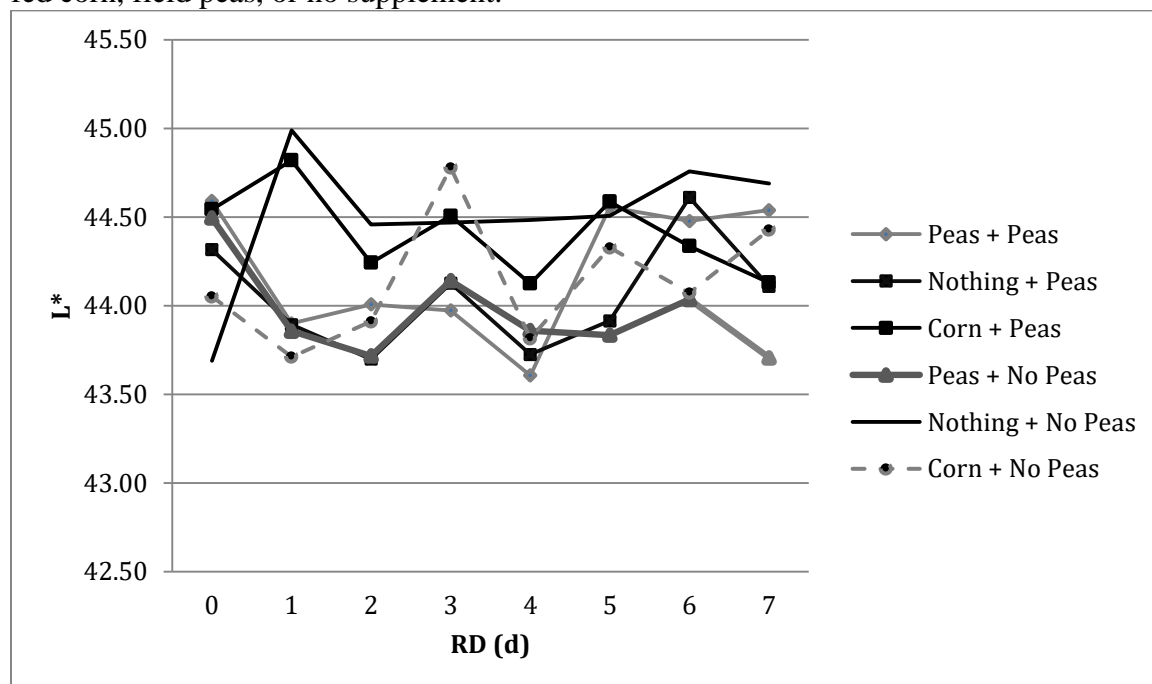
<sup>2</sup>There were no differences among treatments ( $P > 0.05$ ).  $SEM = 0.0690$ .

**APPENDIX X:** Discoloration (%) of strip loin steaks (*L. lumoborum*) aged for 14 d from cattle fed corn, field peas, or no supplement.



Discoloration displayed a retail display by pasture treatment by finishing treatment interaction ( $P < 0.0001$ ;  $SEM=0.3585$ )

**APPENDIX XI:**  $L^*$  values of strip loin steaks (*L lumoborum*) aged for 14 d from cattle fed corn, field peas, or no supplement.



$L^*$  displayed a retail display by pasture treatment by finishing treatment interaction ( $p = 0.0524$ ;  $SEM = 0.4909$ )